Ref #	Hits	Search Query	DBs	Default Operator	Plurals	Time Stamp
L1	20441	vascula$10 near3 (permeab$8 or leak$6) or edema$	US-PGPUB; USPAT	OR	OFF	2004/07/29 08:03
L2	9841	src	US-PGPUB; USPAT	OR	OFF	2004/07/29 08:09
L3	817	1 and 2	US-PGPUB; USPAT	OR	OFF	2004/07/29 08:09
L4	24	1 same 2	US-PGPUB; USPAT	OR	OFF	2004/07/29 08:10
L5	922	2 near10 (inhibit$8 or decreas$8)	US-PGPUB; USPAT	OR	OFF	2004/07/29 08:16
L6	108	1 and 5	US-PGPUB; USPAT	OR	OFF	2004/07/29 08:16
L8	1	"5593997".PN.	USPAT	ADJ	OFF	2004/07/29 08:49

Ref #	Hits	Search Query	DBs	Default Operator	Plurals	Time Stamp
L1	20441	vascula$10 near3 (permeab$8 or leak$6) or edema$	US-PGPUB; USPAT	OR	OFF	2004/07/29 08:03
L2	9841	src	US-PGPUB; USPAT	OR	OFF	2004/07/29 08:09
L3	817	1 and 2	US-PGPUB; USPAT	OR	OFF	2004/07/29 08:09
L4	24	1 same 2	US-PGPUB; USPAT	OR	OFF	2004/07/29 08:10

PGPUB-DOCUMENT-NUMBER: 20040138251

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040138251 A1

TITLE: Thieno[3,2-b]pyridine-6-carbonitriles and thieno[2,3-b]pyridine-5-carbonit- riles as protein kinase inhibitors

PUBLICATION-DATE: July 15, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Boschelli, Diane Harris	New City	NY	US	
Zhang, Nan	Bayside	NY	US	
Barrios Sosa, Ana Carolina	Warwick	NY	US	
Durutlic, Haris	New Windsor	NY	US	
Wu, Biqi	Nanuet	NY	US	

APPL-NO: 10/ 719359

DATE FILED: November 21, 2003

RELATED-US-APPL-DATA:

non-provisional-of-provisional 60428862 20021125 US

US-CL-CURRENT: 514/301, 514/151 , 546/114

ABSTRACT:

This invention provides compounds of Formula (Ia)-(If) 1
wherein:
X, R.sup.1, and R.sup.2 are defined hereinbefore in the specification, which are useful in the treatment of cancer, stroke, osteoporosis, polycystic kidney disease, autoimmune disease, rheumatoid arthritis, and transplant rejection and process for producing said compounds.

[0001] This application claims priority from copending provisional application Serial No. 60/428,862 filed on Nov. 25, 2002 the entire disclosure of which is hereby incorporated by reference.

---------- KWIC ---------

Summary of Invention Paragraph - BSTX (5):
[0005] Src inhibitors may prevent the secondary injury that results from a VEGF-mediated increase in **vascular permeability** such as that seen following stroke [Eliceiri, B. P., Mol. Cell., 4, 915 (1999); Paul, R., Nat. Med. 7, 222 (2001)].

PGPUB-DOCUMENT-NUMBER: 20040038207

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040038207 A1

TITLE: Gene expression in bladder tumors

PUBLICATION-DATE: February 26, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Orntoft, Torben F.	Aabyhoj		DK	

APPL-NO: 09/ 951968

DATE FILED: September 14, 2001

RELATED-US-APPL-DATA:

child 09951968 A1 20010914

parent division-of 09510643 20000222 US UNKNOWN

US-CL-CURRENT: 435/6

ABSTRACT:

Methods for analyzing tumor cells, particularly bladder tumor cells employ gene expression analysis of samples. Gene expression patterns are formed and compared to reference patterns. Alternatively gene expression patterns are manipulated to exclude genes which are expressed in contaminating cell populations. Another alternative employs subtraction of the expression of genes which are expressed in contaminating cell types. These methods provide improved accuracy as well as alternative basis for analysis from diagnostic and prognostic tools currently available.

[0001] This application claims the benefit of U.S. Provisional Application No. 60/121,124, filed Feb. 22, 1999, which is hereby incorporated by reference in its entirety.

---------- KWIC ---------

Detail Description Table CWU - DETL (118):
pos M10612_at HUMAPOCII apolipoprotein C-II gene, av dif pos M11119_at HUMERRNA endogenous retrovirus envelope av dif pos M11147_at HUMFERI ferritin L chain mRNA, ;ferri av dif pos M11313_s_at HUMA2M alpha-2-macroglobulin mRNA, : av dif pos M11353_at HUMHISH3C H3.3 histone class C mRNA, av dif pos M12529_at HUMAPOE apolipoprotein E mRNA, :apoli av dif pos M12529_at HUMAPOE apolipoprotein E mRNA, :apoli av dif pos M12886_at HUMTCBYY T-cell receptor active beta-ch av dif pos M12886_at HUMTCBYY T-cell receptor active beta-ch av dif pos M13207_at HUMCSFGMA granulocyte-macrophage colony- av dif pos M13580_s_at HUMIAIG6 Ia-associated invariant gamma- av dif pos M13666_at HUMCMYBB c-myb mRNA, 3'end. av dif pos M13755_at HUMIFN15K interferon-induced 17-kDa/15-k av dif pos M13829_s_at HUMPKS putative raf related protein av dif

pos M13829_s_at HUMPKS putative raf related protein av dif pos M13903_at HUMINV2 involucrin gene, exon 2. :invo av dif pos M13929_s_at HUMMYCPOA c-myc-P64 mRNA, initiating fro av dif pos M13929_s_at HUMMYCPOA c-myc-P64 mRNA, initiating fro av dif pos M13934_cds2_at HUMRPS14 ribosomal protein S14 gene, av dif pos M13955_at HUMKERMII mesothelial keratin K7 \(type av dif pos M14199_s_at HUMLAMR laminin receptor \(2H5 epitope av dif pos M14199_s_at HUMLAMR laminin receptor \(2H5 epitope av dif pos M14328_s_at HUMENOA alpha enolase mRNA, :enolase av dif pos M14483_ma1_s_at HUMTHYMAA prothymosin alpha mRNA, av dif pos M14676_at HUMSLK src-like kinase \(slk\) mRNA, av dif pos M14876_at HUMSLK src-like kinase \(slk\) mRNA, av dif pos M15395_at HUMLAP leukocyte adhesion protein \(L av dif pos M15661_at HUMRPZH21 ribosomal protein mRNA, :ribo av dif pos M15661_at HUMRPZH21 ribosomal protein mRNA, :ribo av dif pos M16038_at HUMLYN lyn mRNA encoding a tyrosine k av dif pos M17733_at HUMTHYB4 thymosin beta-4 mRNA, :thymos av dif pos M17863_s_at HUMFFI2B preproinsulin-like growth fact av dif pos M17863_s_at HUMFFI2B preproinsulin-like growth fact av dif pos M17885_at HUMPPARP0 acidic ribosomal phosphoprotei av dif pos M17886_at HUMPPARP1 acidic ribosomal phosphoprotei av dif pos M18000_at HUMRPS17A ribosomal protein S17 gene, av dif pos M18737_ma1_at HUMHFSP Hanukah factor senne protease av dif pos M19045_f_at HUMLSZH lysozyme mRNA, :"lysozyme""m av dif pos M19159_at HUMALPPD placental heat-stable alkaline av dif pos M19159_at HUMALPPD placental heat-stable alkaline av dif pos M19301_at HUMKAD brandied-chain aipha-keto acid av dif pos M19878_s_at HUMCALB01 calbindin 27 gene, exons 1 and av dif pos M20902_at HUMAPOCIA apolipoprotein C-I \(VLDL\) ge av dif pos M20902_at HUMAPOCIA apolipoprotein C-I \(VLDL\) ge av dif pos M21142_cds2_s_at HUMGNAS6 guanine nucleotide-binding pro av dif pos M21142_cds2_s_at HUMGNAS6 guanine nucleotide-binding pro av dif pos M21186_at HUMNCBLCA neutrophil cytochrome b light av dif pos M21186_at HUMNCBLCA neutrophil cytochrome b light av dif pos M21302_at HUMSPR2B sma1l proline rich protein \(s av dif pos M21984_at HUMTRT \(clone PWHTnT16\) skeletal mu av dif pos M22490_at HUMBMP2B bone morphogenetic protein-2B av dif pos M22960_at HUMPPR protective protein mRNA, :pro av dif pos M23178_s_at HUMGOS19A homologue-1 of gene encoding a av dif pos M23613_at HUMNPM nucleophosmin mRNA, :nucleoph av dif pos M24194_at HUMMHBA123 MHC protein homologous to chic av dif pos M24194_at HUMMHBA123 MHC protein homologous to chic av dif pos M24485_s_at HUMGSTP1G \(clone pHGST-pi\) glutathione av dif pos M24486_s_at HUMPYHBASA prolyl 4-hydroxylase alpha sub av dif pos M25079_s_at HUMBETGLA sickle cell beta-globin mRNA, av dif pos M25079_s_at HUMBETGLA sickle cell beta-globin mRNA, av dif pos M25280_at HUMLNHR lymph node homing receptor mRN av dif pos M26311_s_at HUMCFA cystic fibrosis antigen mRNA, av dif pos M26311_s_at HUMCFA cystic fibrosis antigen mRNA, av dif pos M26665_s_at HUMHIS2X histatin 2 \(HIS2\) mRNA, :hi av dif pos M28708_s_at HUMPTAA prothymosin alpha mRNA \(ProT- av dif pos M26730_s_at HUMQBPC6 mitochondrial ubiquinone-bindi av dif pos M27281_at HUMVPF vascular permeability factor m av dif pos M27749_r_at HUMIGLR141 immunoglobulin-related 14.1 pr av dif pos M27749_r_at HUMIGLR141 immunoglobulin-related 14.1 pr av dif pos M27826_at HUMRTVLH3 endogenous retroviral protease av dif pos M27891_at HUMCYS3A3 cystatin C \(CST3\) gene, exon av dif pos M28212_at HUMRAB6A GTP-binding protein \(RAB6\) m av dit pos M28882_s_at HUMMUC18B MUC18 glycoprotein mRNA, :mel av dif pos M28882_s_at HUMMUC18B MUC18 glycoprotemn mRNA, :mel av dif pos M29335_at HUMMHDOA MHC class II DO-alpha mRNA, av dif pos M29335_at HUMMHDOA MHC class II DO-alpha mRNA, av dif pos M29610_s_at HUMGLYE glycophorin E mRNA, :glycopho av dif pos M30818_at HUMMXB interferon-induced cellular re av dint pos M30938_at HUMKUP Ku \(p70/p80\) subunit mRNA, av dif pos M31303_ma1_at HUMOP18A oncoprotein 18 \(Op18\) gene, av dif pos M31303_ma1_at HUMOP18A oncoprotein 18 \(Op18\) gene, av dif pos M31520_at HUMRPS24A nbosoma1 protein S24 mRNA. av dif pos M31520_ma1_s_at HUMRPS24A ribosomal protein S24 mRNA. :r av dif pos M31627_at HUMHXBP1 X box binding protein-1 \(XBP- av dif pos M31994_at HUMALDC13 aldehyde dehydrogenase \(ALDH1 av dif pos M32053_at HUMH19 H19 RNA gene, av dif pos M32304_s_at

HUMMET metalloproteinase inhibitor mR av dif pas M32405_at HUMRIGA homologue of rat insulinoma ge av dif pos M32886_at HUMSRICPA Saran CP-22 mRNA, :sorcin :s av dif pos M33600_f_at HUMMHDRA1C MHC class II HLA-DR-beta-1 \(H av dif pos M33680_at HUMTAPA1 26-kDa cell surface protein TA av dif pos M33684_s_at HUMPPPB1A5 \(clone lambda-16-1\) non-rece av dif pos M34041_at HUMADRA2RA alpha-2-adrenergic receptor \(av dif pos M34182_at HUMPRKACG testis-specific protein kinase av dif pos M34516_at HUMIGL122 omega light chain protein 14.1 av dif pos M34516_r_at HUMIGL122 omega light chain protein 14.1 av dif pos M34516_r_at HUMIGL122 omega light chain protein 14.1 av dif pos M34715_at HUMPSBGAA pregnancy-specific beta-1-glyc av dif pos M34998_s_at HUMOQA1A MHC cell surface glycoprotein av dif pos M34996_s_at HUMDQA1A MHC cell surface glycoprotein av dif pos M35198_at HUMINTB6A integrin 8-8 mRNA, :integrin, av dif pos M35252_at HUMCOOTAA CO-029. :transmembrane 4 super av dif pos M35878_at HUMIBP3 insulin-like growth factor-bin av dif pos M36072_at HUMRPL7A nbosoma1 protein L7a \(surf 3 av dif pos M37238_s_at HUMPLC phospholipase C mRNA, :phosph av dif pos M37245_at HUMIGCTL3 Ig superfamily cytotoxic T-lym av dif pos M37245_at HUMIGCTL3 Ig superfamily cytotoxic T-lym av dif pos M37435_at HUMCSDF1 macrophage-specific colony-sti av dif pos M37583_at HUMHIS2AZ histone \(H2A.Z\) mRNA, :hist av dit pos M37815_cds1_at HUMCD284 T-cell membrane glycoprotein C av dif pos M38449_s_at HUMTGFBA transforming growth factor-bet av dif pos M38690_at HUMANTCD9 CD9 antigen mRNA, :CD9 antige av dif pos M38890_at HUMANTCD9 CD9 antigen mRNA, :CD9 antige av dif pos M54995_at HUMCTAP3 connective tissue activation p av dif pea M55409_a_at HUMPANCAN pancreatic tumor-related prote av dif pos M55409_s_at HUMPANCAN pancreatic tumor-related prote av dif pos M57293_at HUMPTHRPA parathyroid hormone-related pe av dif pos M57399_at HUMHBNF1 nerve growth factor \(HBNF-1\) av dif pos M57399_at HUMHBNF1 nerve growth factor \(HBNF-1\) av dif pos M57486_s_at HUMMHDPL MHC class II HLA-DP light chai av dif pos M57468_s_at HUMMHDPL MHC class II HLA-DP light chai av dif pos M57710_at HUMBPIGE IgE-binding protein \(epsilon- av dif pos M58378_cds1_at HUMSYN1E13 synapsmn I\(SYN1\) gene, exon av dif pos M58525_s_at HUMCOMTC catechol-O-methyltransferase av dif pos M58525_s_at HUMCOMTC catechol-O-methyltransferaseav dif pos M59216_s_at UMGABRB1S5 gamma-aminobutyric add-A \(GA av dif pos M59371_at HUMECK protein tyrosine kinase mRNA, av dif pos M59807_at HUMNK4 NK4 mRNA, :natural killer cel av dif pos M59830_at HUMMHHSP2 MHC class III HSP70-2 gene \(H av dif pos M59911_at HUMINTA3A integrin alpha-3 chain mRNA, av dif pos M60483_ma1_s_at HUMPP2AA protein phosphatase 2A catalyt av dif pos M60854_at HUMSRAA ribosomal protein S16 mRNA, av dif pos M61916_at HUMLAM101 laminin B1 chain mRNA, :lamin av dif pos M62403_s_at HUMIGFBPS insulin-like growth factor bin av dif pos M62403_s_at HUMIGFBP5 insulin-like growth factor bin av dif pos M62486_at UMPRPC4S12 C4b-binding protein gene, exon av dif pos M63256_at HUMCDR2AA major Yo paraneoplastic antige av dif pos M63379_at HUMTRPM2A4 TRPM-2 protein gene, exons 7,8 av dif pos M63438_s_at HUMIGGK Ig rearranged gamma chain mRNA av dif pos M63438_s_at HUMIGGK Ig rearranged gamma chain mRNA av dif pos M63573_at HUMSCYLP secreted cyclophilin-like prot av dif pos M63589_at HUMSCL7 stem cell leukemia gene produc log neg M64347_at HUMFGFLR novel growth factor receptor m log neg M64347_at HUMFGFLR novel growth factor receptor m log neg M64873_at HUMHSF1 heat shock factor 1 \(TCF5\) m log neg M64716_at HUMRPS25 ribosomal protein S25 mRNA, : log neg M64992_at HUMPROS30 prosomal protein P30-33K \(pro log neg M65292_s_at HUMHAAA factor H homologue mRNA, :"fa log neg M65292_s_at HUMHAAA factor H homologue mRNA, :"fa log neg M69023_at HUMGGEFERA globin gene. log neg M69066_at HUMMOESIN moesin mRNA, :moesin :moesin log neg M69238_at HUMARNTA aryl hydrocarbon receptor nucl log neg M73077_at HUMGRF1A glucocorticoid receptor repres log neg M73239_s_at HUMSCFA1 \(clone SF1\) hepatocyte growt log neg M73547_at HUMPOLLA polyposis locus \(DP1 gene\) m log neg M74093_at HUMCLNC cyclin mRNA. :cyclin E1 log neg M74297_at HUMHOX14 homeobox 1.4 protein mRNA, :h log neg M74715_s_at HUMIDNAL alpha-L-iduronidas \(IDUA\) mR log neg M77232_ma1_at HUMRPS6B

ribosomal protein S6 gene, com log neg M77836_at HUMP5CR pyrroline 5-carboxylate reduct log neg M80244_at HUME16GEN E18 mRNA, log neg M80254_at HUMCYP cyclophilin isoform \(hCyP3\) log neg M80359_at HUMP78A protein p78 mRNA, :MAP/microt log neg M80563_at HUMCAPL CAPL protein mRNA, :S100 calc log neg M80563_at HUMCAPL CAPL protein mRNA, :S100 cab log neg M80899_at HUMAHNAKA novel protein AHNAK mRNA, part log neg M81750_at HUMMCNDA myeloid cell nuclear different log neg M81757_at HUMS19RP S19 ribosomal protein mRNA, log neg M81883_at HUMGAD67A glutamate decarboxylase \(GAD6 log neg M83181_at HUMHTRB serotonin receptor gene, :5-h log neg M84424_at HUMCTSE09 cathepsin E \(CTSE\) gene, exo log neg M84711_at HUMFTE1A v-fos transformation effector log neg M85289_at HUMHSPG2B heparan sulfate proteoglycan log neg M86400_at HUMPHPLA2 phospholipase A2 mRNA, :tyros log neg M86699_at HUMTTK kinase \(TTK\) mRNA, :TTK pro log neg

US-PAT-NO: 6764833

DOCUMENT-IDENTIFIER: US 6764833 B1

TITLE: Mutated Src oncogene composition and methods

DATE-ISSUED: July 20, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Yeatman; Timothy J.	Tampa	FL	N/A	N/A
Irby; Rosalyn B.	Tampa	FL	N/A	N/A

APPL-NO: 09/ 444711

DATE FILED: November 24, 1999

US-CL-CURRENT: 435/69.1, 435/320.1 , 435/325 , 530/300 , 530/350 , 536/1.11 , 536/18.7 , 536/22.1 , 536/23.1 , 536/23.2 , 536/23.5

ABSTRACT:

The present invention provides a mutant oligonucleotide composition encoding a cellular c-Src tyrosine kinase oncogene. Methods for isolating, expressing and characterizing recombinant Src mutant polypeptide are also provided. The invention further relates to methods for utilizing such oligonucleotides, polypeptides, agonists and antagonists for applications, which relate to research, diagnostics, and clinical arts. More specifically, this invention provides methods of diagnosing, treating, immunizing, and creating transgenic animals based on use of such mutant Src.

20 Claims, 14 Drawing figures

Exemplary Claim Number: 1

Number of Drawing Sheets: 4

---------- KWIC ---------

Detailed Description Text - DETX (113):

DNA clones for microinjection are cleaved with appropriate restriction enzymes, such as Sal1, Not1, etc., and the DNA fragments electrophoresed on 1% agarose gels in TBE buffer (U.S. Pat. No. 5,811,633). The DNA bands are visualized by staining with ethidium bromide, excised, and placed in dialysis bags containing 0.3M sodium acetate at pH 7.0. The DNA is then electroeluted into the dialysis bags, extracted with phenol-chloroform (1:1), and precipitated by two volumes of ethanol. The DNA is redissolved in 1 ml of low salt buffer (0.2M NaCl, 20 mM Tris, pH 7.4, and 1 mM EDTA) and purified on an Elutip-D column. The column is first primed with 3 ml of high salt buffer (1M NaCl, 20 mM Tris, pH 7.4, and 1 mM EDTA) followed by washing with 5 ml of low salt buffer. The DNA solutions are passed through the column for three times to bind DNA to the column matrix. After one wash with 3 ml of low salt buffer, the DNA is eluted with 0.4 ml of high salt buffer and precipitated by two volumes of ethanol. DNA concentrations are measured by absorption at 260 nm in a UV spectrophotometer. For microinjection, DNA concentrations are adjusted to about 3 .mu.g/ml in 5 mM Tris, pH 7.4 and 0.1 mM EDTA. Other methods for

purification of DNA for microinjection are also known. The purified inserts form pcSrc531RI plasmids are then microinjected into the pronuclei of fertilized (C57BL/6.times.CBA)F2 mouse embryos and surviving embryos are transferred into pseudopregnant females according to standard procedures such as disclosed in U.S. Pat. Nos. 5,877,397, 5,907,078, 5,849,993, 5,602,309, 5,387,742, which are incorporated herein by way of reference. SRC531 construct is operably linked to a suitable promoter, e.g., RSV long terminal repeat (LTR), glial fibrillary acidic protein (GFAP), or human beta-globin promoter (GF). Mice that developed from injected embryos are analyzed for the presence of transgene sequences by Southern blot analysis of mutant DNA. Transgene copy number is estimated by band intensity relative to control standards containing known quantities of cloned DNA. At 3 to 8 weeks of age, cells are isolated from these animals and assayed for the presence of transgene encoded SRC 531 mutation. All of the control non-transgenic mice tested negative for expression of SRC 531. Southern blot analysis indicates that many of these mice contain one or more copies of the transgene per somatic and/or germ cell. Some mice with high levels of Src expression developed abnormally, including edemas, head deformities, eye, axial system defects and usually these mice did not survive. Surviving transgenic mice exhibit malignant and/or benign transformation early in their life. Tumors include lymphomas, thymomas, fibrosarcomas, angiosarcomas, hemangiomas, neurofibrosarcomas, etc. These mice are useful as a model for studying SRC 531 mutants in vivo for testing, for example, drugs or SRC 531 antagonists.

US-PAT-NO: 6713462

DOCUMENT-IDENTIFIER: US 6713462 B2

TITLE: Quinolinones and uses thereof

DATE-ISSUED: March 30, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Metcalf, III; Chester A.	Needham	MA	N/A	N/A
Shakespeare; William C.	Southborough	MA	N/A	N/A
Sawyer; Tomi K.	Southborough	MA	N/A	N/A
Wang; Yihan	Newton	MA	N/A	N/A
Bohacek; Regine	Boston	MA	N/A	N/A
Sundaramoorthi; Rajeswari	Watertown	MA	N/A	N/A

APPL-NO: 10/ 177500

DATE FILED: June 21, 2002

PARENT-CASE:

PRIORITY INFORMATION

The present application claims priority under 35 U.S.C. .sctn.119 to U.S. provisional application No. 60/299,936, filed Jun. 21, 2001, entitled "Novel Quinolinones and Uses Thereof", the entire contents of which are hereby incorporated by reference.

US-CL-CURRENT: 514/82, 514/312 , 546/153 , 546/155 , 546/157 , 546/158 , 546/23

ABSTRACT:

The invention relates to compounds of the general formula (and pharmaceutically acceptable derivatives thereof): ##STR1## in which R.sup.A, R.sup.B, R.sup.C, R.sup.D, R.sup.5, R.sup.7, R.sup.9, R.sup.9a, AK, p, q, r and X are as defined herein, and to their preparation and use.

75 Claims, 0 Drawing figures

Exemplary Claim Number: 1

---------- KWIC --------

Brief Summary Text - BSTX (7):

Protein kinases, specifically Src protein kinases, have been shown to play a crucial role in osteoclast function and thus in the resorption of bone and the progression of the osteoporosis. In addition, cellular signal transduction mediated by kinases like Src is believed to play a key role in other diseases, for example cancer and diseases involving increased vascular permeability. Though the exact mechanisms of signal transduction is still unclear, tyrosine kinases have been shown to be important contributing factors in cell proliferation, carcinogenesis and cell differentiation.

Brief Summary Text - BSTX (121):

This invention provides a new family of compounds with a range of biological properties. Compounds of this invention have biological activities relevant for the treatment of diseases including bone related disorders, disorders related to cellular proliferation (e.g., cancer) and disorders related to increased vascular permeability and/or angiogenesis. More generally, the compounds are useful in the regulation of signal transduction pathways. For example, certain compounds of the invention are useful for inhibiting tyrosine kinases, including without limitation receptor-type tyrosine kinases such as those of the HER (e.g. EGFR, HER2, HER3 and HER4), PDGF and FLK families (including, e.g., VEGF-R1 and VEGF-R2) as well as non-reecptor-type tyrosine kinases such as those of the Src and abl subfamilies, again as non-limiting examples.

Brief Summary Text - BSTX (178):

In yet another embodiment, in addition to the treatment or prevention of osteoporosis or cancer, the present invention can be utilized to inhibit increases in vascular permeability. For example, certain compounds are tested for the ability to inhibit the tyrosine kinase activity associated with the VEGF receptors such as Flt and/or KDR and for their ability to inhibit angiogenesis and/or increased vascular permeability. Additionally, these compounds can be tested for the ability to inhibit the tyrosine kinase activity associated with Src and for their ability to inhibit angiogenesis and/or increased vascular permeability. These properties may be assessed, for example, using one or more of the procedures set out below. Thus according to this aspect of the invention there is provided a method for reducing vascular permability in a subject comprising administering a compound of Formula I, as described herein and as described by the various classes and subclasses.

Detailed Description Text - DETX (158):

In addition to their ability to inhibit bone resorption, the compounds of the present invention are also able to inhibit protein kinase activity. For example, inventive compounds can be assessed for their ability to inhibit the activity of receptor and non-receptor tyrosine protein kinases. For example, the present invention presents a general method for determining the ability inhibit the activity of non-receptor tyrosine protein kinases (e.g., members of the src family, abl kinase, and ZAP70 kinase) and receptor tyrosine protein kinases (e.g., EGF family (c-erbB2, c-erbB3, and c-erbB4), the PDGF family (e.g., PDGF receptor, CSF-1, Kit, VEGF and FGF). Thus, the inventive compounds can be used in the immunomodulation and in the treatment of diseases of the immune system, for example in the case of inflammations or organ transplants. They are also suitable for the treatment of hyperproliferative diseases, including, but not limited to psoriasis, tumors, carcinomas and leukemias, and in fibrosis and restenosis. Additionally, compounds can be utilized for the treatment of diseases of the central or the peripheral nervous system where signal transmission by at least one tyrosine protein kinase is involved. Furthermore, Src and certain other kinases are believed to mediate signaling activity in response to a variety of growth factors, including VEGF, vascular endothelial growth factor, which is is an angiogenic factor that promotes vascular permeability, and thus certain inhibitors are usefulas antiangiogenic agents. In addition to the kinase assays described in this section, certain other kinase assays are described in the context of anti-angiogenic agents below, for example.

Detailed Description Text - DETX (193):

As mentioned above, certian kinases are believed to mediate signaling activity in response to a variety of growth factors, including VEGF, vascular endothelial growth factor, which is an angiogenic factor that promotes vascular permeability. For example, certain compounds are tested for the ability to

inhibit the tyrosine kinase activity associated with the VEGF receptors such as Flt and/or KDR and for their ability to inhibit angiogenesis and/or increased vascular permeability. Additionally, these compounds can be tested for the ability to inhibit the tyrosine kinase activity associated with Src and for their ability to inhibit angiogenesis and/or increased vascular permeability. These properties may be assessed, for example, using one or more of the procedures set out below:

US-PAT-NO: 6706699

DOCUMENT-IDENTIFIER: US 6706699 B2

TITLE: Quinolines and uses thereof

DATE-ISSUED: March 16, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Wang; Yihan	Newton	MA	N/A	N/A
Metcalf, III; Chester A.	Needham	MA	N/A	N/A
Shakespeare; William C.	Southborough	MA	N/A	N/A
Sawyer; Tomi K.	Southborough	MA	N/A	N/A
Bohacek; Regine	Boston	MA	N/A	N/A
Sundaramoorthi; Rajeswari	Watertown	MA	N/A	N/A

APPL-NO: 10/ 177990

DATE FILED: June 21, 2002

PARENT-CASE:

PRIORITY INFORMATION

The present application claims priority under 35 U.S.C. .sctn.119 to U.S. provisional application No. 60/299,918, filed Jun. 21, 2001, entitled "Novel Quinolines and Uses Thereof", the entire contents of which are hereby incorporated by reference.

US-CL-CURRENT: 514/82, 514/312 , 514/313 , 546/153 , 546/159 , 546/162 , 546/23

ABSTRACT:

This invention relates to compounds of the general formula: ##STR1##

in which R.sup.A, R.sup.B, R.sup.C and R.sup.D are as defined herein, and to their preparation and use.

44 Claims, 0 Drawing figures

Exemplary Claim Number: 1

---------- KWIC ---------

Brief Summary Text - BSTX (7):

Protein kinases, specifically Src protein kinases, have been shown to play a crucial role in osteoclast function and thus in the resorption of bone and the progression of the osteoporosis. In addition, cellular signal transduction mediated by kinases like Src is believed to play a key role in other diseases, for example cancer and diseases involving increased vascular permeability. Though the exact mechanisms of signal transduction is still unclear, tyrosine kinases have been shown to be important contributing factors in cell proliferation, carcinogenesis and cell differentiation.

Detailed Description Text - DETX (331):
This invention provides a new family of compounds with a range of biological properties. Compounds of this invention have biological activities relevant for the treatment of diseases including bone related disorders, disorders related to cellular proliferation (e.g., cancer) and disorders related to increased vascular permeability and/or angiogenesis. More generally, the compounds are useful in the regulation of signal transduction pathways. For example, certain compounds of the invention are useful for inhibiting tyrosine kinases, including without limitation receptor-type tyrosine kinases such as those of the HER (e.g. EGFR, HER2, HER3 and HER4), PDGF and FLK families (including, e.g., VEGF-R1 and VEGF-R2) as well as non-reecptor-type tyrosine kinases such as those of the Src and abl subfamilies, again as non-limiting examples.

Detailed Description Text - DETX (390):
In yet another embodiment, in addition to the treatment or prevention of osteoporosis or cancer, the present invention can be utilized to inhibit increases in vascular permeability. For example, certain compounds are tested for the ability to inhibit the tyrosine kinase activity associated with the VEGF receptors such as Flt and/or KDR and for their ability to inhibit angiogenesis and/or increased vascular permeability. Additionally, these compounds can be tested for the ability to inhibit the tyrosine kinase activity associated with Src and for their ability to inhibit angiogenesis and/or increased vascular permeability. These properties may be assessed, for example, using one or more of the procedures set out below. Thus according to this aspect of the invention there is provided a method for reducing vascular permability in a subject comprising administering a compound of Formula I, as described herein and as described by the various classes and subclasses.

Detailed Description Text - DETX (557):
In addition to their ability to inhibit bone resorption, the compounds of this invention are also able to inhibit protein kinase activity. For example, inventive compounds can be assessed for their ability to inhibit the activity of receptor and non-receptor tyrosine protein kinases. For example, the present invention presents a general method for determining the ability inhibit the activity of non-receptor tyrosine protein kinases (e.g., members of the src family, abl kinase, and ZAP70 kinase) and receptor tyrosine protein kinases (e.g., EGF family (c-erbB2, c-erbB3, and c-erbB4), the PDGF family (e.g., PDGF receptor, CSF-1, Kit, VEGF and FGF). Thus, the inventive compounds can be used in the immunomodulation and in the treatment of diseases of the immune system, for example in the case of inflammations or organ transplants. They are also suitable for the treatment of hyperproliferative diseases, including, but not limited to psoriasis, tumors, carcinomas and leukemias, and in fibrosis and restenosis. Additionally, compounds can be utilized for the treatment of diseases of the central or the peripheral nervous system where signal transmission by at least one tyrosine protein kinase is involved. Furthermore, Src and certain other kinases are believed to mediate signaling activity in response to a variety of growth factors, including VEGF, vascular endothelial growth factor, which is is an angiogenic factor that promotes vascular permeability, and thus certain inhibitors are usefulas antiangiogenic agents. In addition to the kinase assays described in this section, certain other kinase assays are described in the context of anti-angiogenic agents below, for example.

Detailed Description Text - DETX (596):
As mentioned above, certian kinases are believed to mediate signaling activity in response to a variety of growth factors, including VEGF, vascular endothelial growth factor, which is an angiogenic factor that promotes vascular permeability. For example, certain compounds are tested for the ability to

inhibit the tyrosine kinase activity associated with the VEGF receptors such as Flt and/or KDR and for their ability to inhibit angiogenesis and/or increased vascular permeability. Additionally, these compounds can be tested for the ability to inhibit the tyrosine kinase activity associated with Src and for their ability to inhibit angiogenesis and/or increased vascular permeability. These properties may be assessed, for example, using one or more of the procedures set out below:

Ref #	Hits	Search Query	DBs	Default Operator	Plurals	Time Stamp
L1	20441	vascula$10 near3 (permeab$8 or leak$6) or edema$	US-PGPUB; USPAT	OR	OFF	2004/07/29 08:03
L2	9841	src	US-PGPUB; USPAT	OR	OFF	2004/07/29 08:09
L3	817	1 and 2	US-PGPUB; USPAT	OR	OFF	2004/07/29 08:09
L4	24	1 same 2	US-PGPUB; USPAT	OR	OFF	2004/07/29 08:10
L5	922	2 near10 (inhibit$8 or decreas$8)	US-PGPUB; USPAT	OR	OFF	2004/07/29 08:16
L6	108	1 and 5	US-PGPUB; USPAT	OR	OFF	2004/07/29 08:16
L8	1	"5593997".PN.	USPAT	ADJ	OFF	2004/07/29 08:49

PGPUB-DOCUMENT-NUMBER: 20040142861

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040142861 A1

TITLE: Compositions comprising conditioned cell culture media and uses thereof

PUBLICATION-DATE: July 22, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Mansbridge, Jonathan N.	La Jolla	CA	US	

APPL-NO: 10/ 165860

DATE FILED: June 7, 2002

RELATED-US-APPL-DATA:

non-provisional-of-provisional 60297177 20010607 US

US-CL-CURRENT: 514/12, 424/94.1 , 424/94.4 , 435/404 , 514/18 , 514/458 , 514/474 , 514/562

ABSTRACT:

The invention relates to compositions comprising cell culture medium conditioned by cells grown in three-dimensional culture. The cells used to condition the medium may be genetically modified to alter the concentration of growth factors and antioxidants in the medium. The conditioned cell medium (conditioned medium) may be used for at least one of cosmetic applications, cosmeceutical applications, and pharmaceutical applications, among other things. The invention also relates to proteins comprising a heterologous sequence that enhances cell penetration. The invention also relates to cells comprising DNA encoding such proteins. Methods for preparing the inventive compounds are also provided.

RELATED APPLICATIONS

[0001] This application claims priority of provisional U.S. Patent Application Serial No. 60/297,177, filed Jun. 7, 2001, which is expressly incorporated herein by reference, in its entirety, for any purpose. This application is related to U.S. patent application Ser. No. 09/313,538, filed May 14, 1999, which is expressly incorporated herein by reference, in its entirety, for any purpose.

---------- KWIC ---------

Detail Description Paragraph - DETX (79):

[0083] Phosphate buffered saline containing 1 mM Na.sub.2EDTA, BHT (10 mg/ml) and SDS was added to the sample. The mixture was vigorously vortexed for 15 min at 4.degree. C. and ethanol was added. Vitamin E was extracted in hexane. Hexane phase was collected and dried under nitrogen. Samples were re-dissolved in vitamin E mobile phase and injected to the HPLC system. Authentic compounds were used to generate standard curves, as described (Sen et

al., Molecular basis of vitamin E action. Tocotrienol potently inhibits glutamate-induced pp60(c-Src) kinase activation and death of HT4 neuronal cells. J Biol. Chem. 2000 Apr. 28;275(17):13049-55; Roy et al., Simultaneous Detection of Tocopherols and Tocotrienols in Biological Samples Using HPLC-Coulometric Electrode Array. Meth. Enzymol. 2001 (in press)). As shown in FIG. 3A, this filtered media preparation comprised 1.62 .mu.M .alpha.-tocopherol and 0.06 .mu.M .gamma.-tocopherol.

Detail Description Paragraph - DETX (81):

[0085] Glutathione (GSH) was extracted from acidified samples and a C-18 column (150 mm.times.4.6 mm, 5 .mu.m pore size; Alltech, Deerfield, Ill.) was used for GSH separation. HPLC was performed as described (Sen et al., Molecular basis of vitamin E action. Tocotrienol potently inhibits glutamate-induced pp60(c-Src) kinase activation and death of HT4 neuronal cells. J Biol. Chem. 2000 Apr. 28;275(17):13049-55). As shown in FIG. 3B, this filtered media preparation contained 3.36 nM GSH.

Detail Description Paragraph - DETX (111):

[0103] Nutrient solution was tested for primary and cumulative irritation on normal, human, adult, forearm skin using standard cosmetic safety protocols. Two hundred microliters of either control or nutrient solution was applied to a 3.8 cm.sup.2 occluded patch (Webril non-woven cotton pad) on the upper forearm. The patch was held in place with a 3M.RTM. hypoallergenic tape. The primary irritation study involved 15 subjects (13 females and two males, 28-77 years of age). Nutrient solution was applied in two 24 hour intervals to the occluded patches on normal and abraided (tape stripped five times using Transpore tape to remove outer layers of the stratum corneum) skin on the subject's upper forearm of. The cumulative irritation study involved 31 subjects (21 females and 10 males, 20-65 years of age). One subject withdrew due to tape irritation and one due to personal reasons. Twenty-nine subjects, 19 females and 10 males completed the study. Nutrient solution was on the upper forearm in 14, consecutive, 24 hour applications. Gross observations were graded for glazing, peeling, scabbing, fissuring, hyperpigmentation and hypopigmentation. Irritation was scored visually using a 5 point scale and graded numerically for erythema, edema, papules, vesicles, bulla reactions, weeping, spreading, and induration. As determined by licensed health care professionals, no adverse events were induced by the nutrient solution or control in these studies.

PGPUB-DOCUMENT-NUMBER: 20040138251

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040138251 A1

TITLE: Thieno[3,2-b]pyridine-6-carbonitriles and thieno[2,3-b]pyridine-5-carbonit- riles as protein kinase inhibitors

PUBLICATION-DATE: July 15, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Boschelli, Diane Harris	New City	NY	US	
Zhang, Nan	Bayside	NY	US	
Barrios Sosa, Ana Carolina	Warwick	NY	US	
Durutlic, Haris	New Windsor	NY	US	
Wu, Biqi	Nanuet	NY	US	

APPL-NO: 10/ 719359

DATE FILED: November 21, 2003

RELATED-US-APPL-DATA:

non-provisional-of-provisional 60428862 20021125 US

US-CL-CURRENT: 514/301, 514/151 , 546/114

ABSTRACT:

This invention provides compounds of Formula (Ia)-(If) 1
wherein:
X, R.sup.1, and R.sup.2 are defined hereinbefore in the specification, which are useful in the treatment of cancer, stroke, osteoporosis, polycystic kidney disease, autoimmune disease, rheumatoid arthritis, and transplant rejection and process for producing said compounds.

[0001] This application claims priority from copending provisional application Serial No. 60/428,862 filed on Nov. 25, 2002 the entire disclosure of which is hereby incorporated by reference.

---------- KWIC ---------

Summary of Invention Paragraph - BSTX (3):
[0003] Tyrosine kinases (TK) are a class of protein kinases. The major family of cytoplasmic protein TKs is the Src family which consists of at least eight members (Src, Fyn, Lyn, Yes, Lck, Fgr, Hck and Blk) that participate in a variety of signaling pathways [Schwartzberg, P. L., Oncogene, 17, 1463 (1998)]. The prototypical member of this tyrosine kinase family is Src, which is involved in proliferation and migration responses in many cell types [Sawyer, T., Expert Opin. Investig. Drugs, 10, 1327 (2001)]. Src activity has been shown to be elevated in breast, colon (.about.90%), pancreatic (>90%) and liver (>90%) tumors. Greatly increased Src activity is also associated with

metastasis (>90%) and poor prognosis. Antisense Src message impedes growth of colon tumor cells in nude mice [Staley, C. A., Cell Growth Differentiation, 8, 269 (1997)], suggesting that Src inhibitors could slow tumor growth. In addition to its role in cell proliferation, Src also acts in stress response pathways, including the hypoxia response. Nude mice studies with colon tumor cells expressing antisense Src message have reduced vascularization [Ellis, L. M., J. Biol. Chem., 273, 1052 (1998)], which suggests that Src inhibitors could be anti-angiogenic as well as anti-proliferative.

Summary of Invention Paragraph - BSTX (4):

[0004] Src disrupts E-cadherin associated cell-cell interactions [E. Avezienyte, Nature Cell Bio., 4, 632 (2002)]. A low molecular weight Src inhibitor prevents this disruption thereby reducing cancer cell metastasis [Nam, J. S., Clinical Cancer Res., 8, 2340 (2002)].

Summary of Invention Paragraph - BSTX (5):

[0005] Src inhibitors may prevent the secondary injury that results from a VEGF-mediated increase in vascular permeability such as that seen following stroke [Eliceiri, B. P., Mol. Cell., 4, 915 (1999); Paul, R., Nat. Med. 7, 222 (2001)].

Summary of Invention Paragraph - BSTX (6):

[0006] Src also plays a role in osteoporosis. Mice genetically engineered to be deficient in Src production were found to exhibit osteopetrosis, the failure to resorb bone [Soriano, P., Cell, 64, 693 (1991); Boyce, B. F., J. Clin., Invest., 90, 1622 (1992)]. This defect was characterized by a lack of osteoclast activity. Since osteoclasts normally express high levels of Src, inhibition of Src kinase activity may be useful in the treatment of osteoporosis [Missbach, M., Bone, 24, 437 (1999)].

PGPUB-DOCUMENT-NUMBER: 20040138189

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040138189 A1

TITLE: Materials and methods for treatment of cancer and identification of anti-cancer compounds

PUBLICATION-DATE: July 15, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Sebti, Said M.	Tampa	FL	US	
Jove, Richard	Tampa	FL	US	

APPL-NO: 10/ 472056

DATE FILED: March 8, 2004

PCT-DATA:
APPL-NO: PCT/US02/11157
DATE-FILED: Mar 28, 2002
PUB-NO:
PUB-DATE:
371-DATE:
102(E)-DATE:

US-CL-CURRENT: 514/172, 514/179

ABSTRACT:

The subject invention pertains to the treatment of tumors and cancerous tissues and the prevention of tumorigenesis and malignant transformation through the modulation of JAK/STAT3 intracellular signaling. The subject invention concerns pharmaceutical compositions containing cucurbitacin I, or a pharmaceutically acceptable salt or analog thereof, to a patient, wherein the tumor is characterized by the constitutive activation of the JAK/STAT3 intracellular signaling pathway. The present invention further pertains to methods of moderating the JAK and/or STAT3 signaling pathwaysin vitro or in vivo using cucurbitacin I, or a pharmaceutically acceptable salt or analog therof. Another aspect of the present invention concerns a method for screening candidate compoudns for JAK AND/or STAT3 inhibition and anti-tumor activity.

CROSS-REFERENCE TO RELATED APPLICATION

[0001] This application claims the benefit of provisional patent application Serial No. 60/279,104, filed Mar. 28, 2001.

---------- KWIC ---------

Brief Description of Drawings Paragraph - DRTX (6):

[0020] FIGS. 5A-5C show the effects of JSI-124 on phosphotyrosine levels and kinase activities of JAK and Src kinases. FIG. 5A shows suppression of

phosphotyrosine levels of STAT3 and JAK2 but not Src, by JSI-124. A549 and MDA-MB-468 cells were treated with various concentrations of JSI-124 and processed for immunoblotting with antibodies specific for either phosphotyrosine STAT3, phosphotyrosine JAK2 or phosphotyrosine Src as described under Methods. The membranes were also reblotted with antibodies to STAT3 and JAK2. FIG. 5B shows rapid suppression by JSI-124 of phosphotyrosine STAT3 and JAK2. A549 and MDA-MB-468 cells were treated with JSI-124 (10 .mu.M) for various lengths of time (0-240 min) and processed as described above. FIG. 5C shows that JSI-124 does not inhibit JAK1, JAK2 and Src kinase activities. Lysates from v-Src transformed cells, A549 cells and MDA-MB-468 cells were immunoprecipitated with antibodies against JAK1, JAK2 and Src. Autophosphorylation kinase assays were then performed as described under Materials and Methods. Immunoprecipitates were incubated either with vehicle control (C), JSI-124 (J), the JAK kinase inhibitor AG490 (A) or the Src kinase inhibitor PD180970 (P). Data are representative of three independent experiments.

Detail Description Paragraph - DETX (9):

[0039] The ability of JSI-124 to suppress the cellular levels of phosphotyrosine-STAT3 but not phospho-Erk1/2, phospho-JNK, and phospho-Akt suggested that a STAT3 tyrosine kinase is a possible molecular target for JSI-124. Consistent with a direct inhibition of the enzymatic activity of a tyrosine kinase is the fact that suppression of the STAT3 phosphotyrosine levels was rapid (observed as early as 30 min and complete after only 2 hr of treatment). There are two well-characterized STAT3 tyrosine kinases: JAK and Src kinase. Because phosphotyrosine JAK2 levels were also reduced by JSI-124 this suggested that JAK2 is likely not the target. This was confirmed by in vitro kinase assays where JAK2 and JAK1 enzymatic activities were inhibited by AG490, a known JAK inhibitor, but not JSI-124. Similarly, Src kinase activity was inhibited in vitro by the known Src kinase inhibitor PD180970 but not JSI-124, indicating that Src kinase is not a target. The receptor tyrosine kinase EGFR that is believed also to phosphorylate STAT3 is most likely not a target either since EGF-stimulation of EGFR tyrosine phosphorylation in the breast cell line MCF-10A and EGFR-overexpressing NIH 3T3 cells was inhibited only minimally by JSI-124 (data not shown).

Detail Description Paragraph - DETX (67):

[0097] Transfection and generation of stable clones. NIH 3T3/v-Src/pLucTKS3 and NIH 3T3/v-Src/pRLSRE are stable clones that were generated by transfecting NIH 3T3/v-Src fibroblasts with pLucTKS3 or pRLSRE and selecting for G418-resisitant and zeocin clones, respectively (Turkson, J. et al. Mol. Cell Biol., 1999, 19:7519-7528; Turkson, J. et al. J. Biol. Chem., 2001, 28:28). In the case of NIH 3T3/v-Src/pLucTKS3/pRLSRE, pRLSRE was transfected into NIH 3T3/v-Src/pLucTKS3 cells and stable G418-resistant clones were selected. Transfections were carried out with LipofectAMINE Plus (Invitrogen Corporation, Carlsbad, Calif.) according to the manufacturer's protocol. Treatment of cells with inhibitors: Src-transformed NIH 3T3 cells stably expressing reporter constructs pLucTKS3 or pRLSRE or both were treated with JSI-124 (10 .mu.M) for 24-48 hr prior to harvesting cells for cytosolic and nuclear extracts preparation and luciferase assay.

Detail Description Paragraph - DETX (78):

[0106] The results of the cytoblot shown in FIG. 2 identifies JSI-124 as an inhibitor of v-Src activation of STAT3 in NIH 3T3 cells. To determine whether JSI-124 suppresses phosphotyrosine STAT3 levels in human cancer cell lines, several human cancer cell lines were evaluated and those with high levels of tyrosine phosphorylated STAT3 were identified, as shown in FIG. 3A. Among the human cancer cell lines evaluated A549 (a lung adenocarcinoma), MDA-MB-468 and MDA-MB-231 (two breast carcinomas), and Panc-1 (a pancreatic carcinoma)

contained high levels of tyrosine phosphorylated STAT3. These human cancer cell lines along with the positive control cell line (v-Src transformed NIH 3T3 cells) were treated with either vehicle or JSI-124 (10 .mu.M) for 4 hr and the cell lysates processed for Western blotting with anti-phosphotyrosine STAT3 antibody as described under Methods. FIG. 3B shows that JSI-124 was very effective at reducing the levels of tyrosine phosphorylated STAT3. These results confirm those of the cytoblot and demonstrate the ability of JSI-124 to suppress the levels of constitutively-activated, tyrosine phosphorylated STAT3 not only in v-Src transformed NIH 3T3 murine fibroblasts but also in human cancer cells of epithelial origin.

Detail Description Paragraph - DETX (86):

[0112] The ability of JSI-124 to suppress phosphotyrosine levels of STAT3 suggests that this agent may interfere with the function of the upstream tyrosine kinases JAK and Src that are known to phosphorylate STAT3. The effects of JSI-124 on the phosphotyrosine levels of JAK2 and Src in whole cells as well as the ability of JSI-124 to inhibit Src, JAK1, and JAK2 kinase activities in vitro were evaluated. FIG. 5A shows that treatment of A549 and MDA-MB-468 with JSI-124 results in reduction of the levels of tyrosine phosphorylated STAT3, with A549 cells being more sensitive than MDA-MB-468. Furthermore, JSI-124 was also effective at suppressing the levels of tyrosine phosphorylated JAK2 but not those of tyrosine phosphorylated Src. JSI-124 had no effect on the protein levels of STAT3 and JAK2 in both cell lines, as shown in FIG. 5A.

Detail Description Paragraph - DETX (88):

[0114] The ability of JSI-124 to inhibit the kinase activities of Src, JAK1, and JAK2 in vitro was next evaluated. To this end, Src, JAK1, and JAK2 were immunoprecipitated from either A549, MDA-MB-468, or v-Src/NIH 3T3 cells and incubated the immunoprecipitates with either vehicle control, JSI-124, the JAK tyrosine kinase inhibitor AG490, or the Src kinase inhibitor PD180970 and followed autophosphorylation of Src, JAK1, and JAK2 as described under Methods. FIG. 5C shows that in all three cell lines as expected PD180970 inhibits Src but not JAK1 or JAK2 activities (A549 did not have JAK1 kinase activity). Similarly, AG490 inhibited JAK1 and JAK2 but not Src kinase activities. In contrast, all three kinase activities were not affected by JSI-124, as shown in FIG. 5C. Therefore, although in whole cells JSI-124 is very effective at suppressing the levels of tyrosine phosphorylated STAT3 and JAK2, it is unable to inhibit directly Src, JAK1, or JAK2 kinase activities in vitro.

Detail Description Paragraph - DETX (94):

[0118] Previous studies have shown that interfering with STAT3 signaling using a gene therapy approach with a dominant negative variant of STAT3 (STAT3-.beta.) resulted in inhibition of the growth of melanoma cells in nude mice (Niu, G. et al. Cancer Res., 1999, 59:5059-5063; Catlett-Falcone, R. et al. Immunity, 1999, 10:105-115). Because JSI-124 inhibits aberrantly activated STAT3 signaling, DNA binding, and STAT3-mediated gene expression, it was reasoned that the growth in nude mice of tumors with constitutively-activated STAT3 should be more sensitive to JSI-124 than that of tumors with low or without constitutively-activated STAT3. To this end, A549 and MDA-MB-468 as well as Calu-1 cells, a lung adenocarcinoma which has barely detectable levels of tyrosine phosphorylated STAT3 (FIG. 3A), were implanted subcutaneously in nude mice. When the tumors reached an average size of about 150 mm.sup.3, the animals were randomized and treated intraperitoneally with either vehicle or JSI-124 (1 mg/kg/day) as described under Methods. FIGS. 7B and 7C, respectively, show that A549 and Calu-1 tumors from animals treated with vehicle grew to about 500 mm.sup.3 twenty-six days after tumor implantation. MDA-MB-468 treated with vehicle control grew to about 300 mm.sup.3 sixty days after tumor implantation, as shown in FIG. 7A. FIGS. 7B and 7A show that

JSI-124 inhibited A549 and MDA-MB-468 tumor growth by 76% and 86%, respectively. In contrast, JSI-124 had little effect on the growth in nude mice of Calu-1 cells, as shown in FIG. 7C. Treatment of mice bearing A549 cells with a reduced dose of 0.5 mg/kg/day for 23 days also inhibited tumor growth by 52% (data not shown). At both doses, 1 mg/kg/day and 0.5 mg/kg/day, JSI-124 had no effects on body weight, activity or food intake of mice. However, at the local site of drug injection, the peritoneal cavity, JSI-124 at the 1 mg/kg/day dose, caused edema. A similar observation was made by the National Cancer Institute Developmental Therapeutics Program where edema was observed at the subcutaneous site of injection (Jill Johnson, NCI, personal communication).

Detail Description Paragraph - DETX (95):

[0119] The results from A549, Calu-1, and MDA-MB-468 xenograft studies suggest that human cancer cells which express constitutively-activated STAT3 should be sensitive to JSI-124. It was further reasoned that if the ability of JSI-124 to inhibit tumor growth in nude mice depends on constitutively-activated STAT3, v-Src-transformed NIH 3T3 cells which require constitutively-activated STAT3 for malignant transformation (Yu, C. L. et al. Science, 1995, 269:81-83) should be sensitive to JSI-124 whereas oncogenic Ras-transformed NIH 3T3 cells, where STAT3 is not constitutively-activated (see FIG. 3A), should be resistant. FIGS. 7D and 7F show that, in the absence of JSI-124, the growth of both v-Src- and Ras-transformed NIH 3T3 tumors was highly aggressive and reached average sizes of about 2500 mm.sup.3 and 1000 mm.sup.3, respectively, within 9 days of tumor cell implantation. FIG. 7D also shows that JSI-124 (1 mg/kg/day) inhibited the growth of v-Src/NIH 3T3 tumors by 64%. In contrast, the growth of Ras/NIH 3T3 tumors was resistant to JSI-124, as shown in FIG. 7F. These results coupled with those from the human tumor xenografts show that JSI-124 selectively targets tumors with constitutively-activated STAT3 signaling.

Detail Description Paragraph - DETX (106):

[0129] The ability of the cucurbitacin analogs (at 1 mg/Kg/day intraperitonealy) to inhibit the growth of v-src transformed NIH 3T3 cells implanted subcutaneously in nude mice was evaluated, as described with respect to cucurbitacin I. FIG. 17 shows that cucurbitacin Q, tetrahydro-cucurbitacin I, cucurbitacin I, and cucurbitacin E, each inhibit tumor growth. Cucurbitacin A inhibited tumor growth to a lesser extent (160%). Cucurbitacin B was toxic at 1 mpk. However, at 0.2 mpk/day, cucurbitacin B was not toxic and inhibited tumor growth by 40%. Next, the ability of the analogs to inhibit phospho STAT3 or phospho JAK2 levels was correlated to their ability to inhibit tumor growth in nude mice. FIG. 16 shows that cucurbitacin I, which inhibits both phospho STAT3 and phospho JAK2 levels (see inset), inhibit tumor growth. In contrast, cucurbitacin A, which inhibit only phospho JAK2 but not phospho STAT3 levels (see inset), did not significantly inhibit tumor growth. Furthermore, cucurbitacin Q, which inhibits phospho STAT3 but not phospho JAK2 levels (see inset), was as potent as curcubatacin I at inhibiting tumor growth. These results indicate that a cucurbitacin with the ability to inhibit STAT3 activation alone (cucurbitacin Q) is sufficient to inhibit tumor growth. However, cucurbitacin A, which inhibits only JAK2 activation, inhibits tumor growth to a lesser extent. Also, the ability of cucurbitacin I to inhibit both JAK2 and STAT3 activation does not make it a better inhibitor of tumor growth than the cucurbitacin that inhibits STAT3 alone. This indicates that, in a model where both JAK2 and STAT3 are actively signaling cells to grow and survive, it may be more important to shut down the constitutive signaling of STAT3.

PGPUB-DOCUMENT-NUMBER: 20040127884

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040127884 A1

TITLE: Vascular irrigation solution and method for inhibition of pain, inflammation, spasm and restenosis

PUBLICATION-DATE: July 1, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Demopulos, Gregory A.	Mercer Island	WA	US	
Palmer, Pamela Pierce	San Francisco	CA	US	
Herz, Jeffrey M.	Mill Creek	WA	US	

APPL-NO: 10/ 674290

DATE FILED: September 29, 2003

RELATED-US-APPL-DATA:

child 10674290 A1 20030929

parent continuation-of 10195625 20020712 US GRANTED

parent-patent 6645168 US

child 10195625 20020712 US

parent continuation-of 09837141 20010417 US GRANTED

parent-patent 6420432 US

child 09837141 20010417 US

parent continuation-of 09072913 19980504 US GRANTED

parent-patent 6261279 US

child 09072913 19980504 US

parent continuation-of 08670699 19960626 US GRANTED

parent-patent 5820583 US

child 08670699 19960626 US

parent continuation-in-part-of PCT/US95/16028 19951212 US PENDING

child PCT/US95/16028 19951212 US

parent continuation-in-part-of 08353775 19941212 US ABANDONED

US-CL-CURRENT: 604/500

ABSTRACT:

A method and solution for perioperatively inhibiting a variety of pain, inflammation, spasm and restenosis processes resulting from cardiovascular or general vascular therapeutic and diagnostic procedures. The solution preferably includes multiple pain and inflammation inhibitory agents and spasm inhibitory agents at dilute concentration in a physiologic carrier, such as saline or lactated Ringer's solution. Specific preferred embodiments of the solution of the present invention for use in cardiovascular and general vascular procedures also include anti-restenosis agents. The solution is introduced luminally to continuously irrigate an arterial site during an operative/interventional or diagnostic procedure for preemptive inhibition of pain and inflammation, vascular and non-vascular smooth muscle spasm, and restenosis while avoiding undesirable side effects associated with oral, intramuscular, subcutaneous or intravenous application of larger doses of the agents. One preferred solution to inhibit pain, inflammation, vasospasm and restenosis includes a serotonin.sub.2 antagonist, a cyclooxygenase inhibitor, an endothelin antagonist, an ATP-sensitive K.sup.+ channel antagonist, a Ca.sup.2+ channel antagonist, a nitric oxide donor, an anti-thrombin agent, a glycoprotein IIb/IIIa receptor blocker, a PKC inhibitor and a protein tyrosine kinase inhibitor.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] The present application is a continuation of U.S. patent application Ser. No. 10/195,625, filed Jul. 12, 2002, which is a continuation of U.S. patent application Ser. No. 09/837,141, filed Apr. 17, 2001, now U.S. Pat. No. 6,420,432, which is a continuation of U.S. patent application Ser. No. 09/072,913, filed May 4, 1998, now U.S. Pat. No. 6,261,279, which is a continuation of U.S. patent application Ser. No. 08/670,699, filed Jun. 26, 1996, now U.S. Pat. No. 5,820,583, which is a continuation-in-part of International Patent Application No. PCT/US95/16028, filed Dec. 12, 1995, designating the United States and which is a continuation-in-part of co-pending U.S. patent application Ser. No. 08/353,775, filed Dec. 12, 1994, now abandoned, priority of the filing dates of which is hereby claimed under 35 U.S.C. .sctn. 120.

---------- KWIC ---------

Summary of Invention Paragraph - BSTX (19):

[0017] Specific preferred embodiments of the solution of the present invention for use in cardiovascular and general vascular procedures include anti-restenosis agents, which most preferably are used in combination with anti-spasm agents. Suitable anti-restenosis agents include: (1) antiplatelet agents including: (a) thrombin inhibitors and receptor antagonists, (b) adenosine disphosphate (ADP) receptor antagonists (also known as purinoceptor.sub.1 receptor antagonists), (c) thromboxane inhibitors and receptor antagonists and (d) platelet membrane glycoprotein receptor antagonists; (2) inhibitors of cell adhesion molecules, including (a) selectin inhibitors and (b) integrin inhibitors; (3) anti-chemotactic agents; (4) interleukin receptor antagonists (which also serve as anti-pain/anti-inflammation agents); and (5) intracellular signaling inhibitors including: (a) protein kinase C (PKC) inhibitors and protein tyrosine kinase inhibitors, (b) modulators of intracellular protein tyrosine phosphatases, (c) inhibitors of src homology.sub.2 (SH2) domains, and (d) calcium channel antagonists. Such agents are useful in preventing restenosis of arteries treated by angioplasty, rotational atherectomy or other cardiovascular or general vascular therapeutic or diagnostic procedures.

Detail Description Paragraph - DETX (5):
[0041] Specific preferred embodiments of the solution of the present invention for use in cardiovascular and general vascular procedures include anti-restenosis agents, which most preferably are used in combination with anti-spasm agents. Suitable anti-restenosis agents include: (1) antiplatelet agents including: (a) thrombin inhibitors and receptor antagonists, (b) adenosine disphosphate (ADP) receptor antagonists (also known as purinoceptor.sub.1 receptor antagonists), (c) ihromboxane inhibitors and receptor antagonists and (d) platelet membrane glycoprotein receptor antagonists; (2) inhibitors of cell adhesion molecules, including (a) selectin inhibitors and (b) integrin inhibitors; (3) anti-chemotactic agents; (4) interleukin receptor antagonists (which also serve as anti-pain/anti-inflammation agents); and (5) intracellular signaling inhibitors including: (a) protein kinase C (PKC) inhibitors and protein tyrosine phosphatases, (b) modulators of intracellular protein tyrosine kinase inhibitors, (c) inhibitors of src homology.sub.2 (SH2) domains, and (d) calcium channel antagonists. Such agents are useful in preventing restenosis of arteries treated by angioplasty, rotational atherectomy or other cardiovascular or general vascular therapeutic procedure.

Detail Description Paragraph - DETX (37):
[0071] Calcitonin gene-related peptide (CGRP) is a peptide which is also colocalized in sensory neurons with substance P, and which acts as a vasodilator and potentiates the actions of substance P. Brain, S. D., et al., Inflammatory Oedema Induced by Synergism Between Calcitonin Gene-Related Peptide (CGRP) and Mediators of Increased Vascular Permeability, Br. J. Pharmacol. 99, p. 202 (1985). An example of a suitable CGRP receptor antagonist is I-CGRP-(8-37), a truncated version of CGRP. This polypeptide inhibits the activation of CGRP receptors. Suitable concentrations for this agent are provided in Table 8.

Detail Description Paragraph - DETX (116):
[0130] The platelet-derived growth factor (PDGF) receptor is of great interest as a target for inhibition in the cardiovascular field since it is believed to play a significant role both in atherosclerosis and restenosis. The release of PDGF by platelets at damaged surfaces of endothelium within blood vessels results in stimulation of PDGF receptors on vascular smooth muscle cells. As described above, this initiates a sequence of intracellular events leading to enhanced proliferation and neointimal thickening. An inhibitor of PDGF kinase activity would be expected to prevent proliferation and enhance the probability of success following cardiovascular and general vascular procedures. Any of several related tyrphostin compounds have potential as specific inhibitors of PDGF-receptor tyrosine kinase activity (IC.sub.50s in vitro in the 0.5-1.0 .mu.M range), since they have little effect on other protein kinases and other signal transduction systems. To date, only a few of the many tyrphostin compounds are commercially available, and suitable concentrations for these agents as used in the present invention are set forth below. In addition, staurosporine has been reported to demonstrate potent inhibitory effects against several protein tyrosine kinases of the src subfamily and a suitable concentration for this agent as used in the present invention also is set forth below.

Detail Description Paragraph - DETX (120):
c. Inhibitors of SH2 Domains (src Homolgy.sub.2 Domains).

Detail Description Paragraph - DETX (147):
[0151] A suitable irrigation solution for control of pain and edema during such arthroscopic techniques is provided in Example I herein below. For arthroscopy, it is preferred that the solution include a combination, and

preferably all, or any of the following: a serotonin.sub.2 receptor antagonist, a serotonin.sub.3 receptor antagonist, a histamine.sub.1 receptor antagonist, a serotonin receptor agonist acting on the 1A, 1B, 1D, 1F and/or 1E receptors, a bradykinin.sub.1 receptor antagonist, a bradykinin.sub.2 receptor antagonist, and a cyclooxygenase inhibitor.

Detail Description Paragraph - DETX (154):

[0158] The solution of the present invention also has utility for reducing pain and inflammation associated with urologic procedures, such as trans-urethral prostate resection and similar urologic procedures. References herein to application of solution to the urinary tract or to the urological structures is intended to include application to the urinary tract per se, bladder and prostate and associated structures. Studies have demonstrated that serotonin, histamine and bradykinin produce inflammation in lower urinary tract tissues. Schwartz, M. M., et al., Vascular Leakage in the Kidney and Lower Urinary Tract: Effects of Histamine, Serotonin and Bradykinin, Proc Soc Exp Biol Med 140, pp. 535-539 (1972). A suitable irrigation solution for urologic procedures is disclosed in Example III herein below. The solution preferably includes a combination, and preferably all, of the following: a histamine.sub.1 receptor antagonist to inhibit histamine-induced pain and inflammation; a 5-HT.sub.3 receptor antagonist to block activation of these receptors on peripheral C-fiber nociceptive neurons; a bradykinin.sub.1 antagonist; a bradykinin.sub.2 antagonist; and a cyclooxygenase inhibitor to decrease pain/inflammation produced by prostaglandins at the tissue injury sites. Preferably an anti-spasm agent is also included to prevent spasm in the urethral canal and bladder wall.

Claims Text - CLTX (7):

6. The method of claim 1, wherein the restenosis inhibitory agent is selected from the group consisting of: antiplatelet agents including thrombin inhibitors and receptor antagonists, purinoceptor antagonists, thromboxane inhibitors and receptor antagonists and platelet membrane glycoprotein receptor antagonists; inhibitors of cell adhesion molecules, including selectin inhibitors and integrin inhibitors; anti-chemotactic agents; interleukin receptor antagonists; and intracellular signaling inhibitors including protein kinase C inhibitors and protein tyrosine kinase inhibitors, modulators of intracellular protein tyrosine phosphatases, inhibitors of src homology.sub.2 domains, and calcium channel antagonists.

Claims Text - CLTX (8):

7. The method of claim 1, wherein the restenosis inhibitory agent is selected from the group consisting of: (a) antiplatelet agents selected from the group consisting of (i) direct thrombin inhibitors and receptor antagonists, (ii) purinoceptor receptor antagonists, (iii) thromboxane inhibitors and receptor antagonists and (iv) platelet membrane glycoprotein receptor antagonists; (b) inhibitors of cell adhesion molecules, including (i) selectin inhibitors and (ii) integrin inhibitors; (c) anti-chemotactic agents; (d) interleukin receptor antagonists; and (e) intracellular signaling inhibitors selected from the group consisting of (i) protein kinase C inhibitors and protein tyrosine kinase inhibitors, (ii) modulators of intracellular protein tyrosine phosphatases, and (iii) inhibitors of src homology.sub.2 domains.

Claims Text - CLTX (30):

29. The method of claim 24, wherein the restenosis inhibitory agents are selected from the group consisting of: antiplatelet agents including thrombin inhibitors and receptor antagonists, purinoceptor antagonists, thromboxane inhibitors and receptor antagonists and platelet membrane glycoprotein receptor antagonists; inhibitors of cell adhesion molecules, including selectin

inhibitors and integrin inhibitors; anti-chemotactic agents; interleukin receptor antagonists; and intracellular signaling inhibitors including protein kinase C inhibitors and protein tyrosine kinase inhibitors, modulators of intracellular protein tyrosine phosphatases, inhibitors of src homology.sub.2 domains, and calcium channel antagonists.

Claims Text - CLTX (31):
30. The method of claim 24, wherein the restenosis inhibitory agents are selected from the group consisting of: (a) antiplatelet agents selected from the group consisting of (i) direct thrombin inhibitors and receptor antagonists, (ii) purinoceptor receptor antagonists, (iii) thromboxane inhibitors and receptor antagonists and (iv) platelet membrane glycoprotein receptor antagonists; (b) inhibitors of cell adhesion molecules, including (i) selectin inhibitors and (ii) integrin inhibitors; (c) anti-chemotactic agents; (d) interleukin receptor antagonists; and (e) intracellular signaling inhibitors selected from the group consisting of (i) protein kinase C inhibitors and protein tyrosine kinase inhibitors, (ii) modulators of intracellular protein tyrosine phosphatases, and (iii) inhibitors of src homology.sub.2 domains.

Claims Text - CLTX (41):
40. A solution for use in the preemptive inhibition of restenosis, and selectively for preemptively inhibiting pain/inflammation and/or spasm, during a vascular procedure, comprising a plurality of agents selected from the group consisting of pain/inflammation inhibitory agents, spasm inhibitory agents and restenosis inhibitory agents in a liquid carrier, the agents being selected to act on a plurality of differing molecular targets, the solution including at least one restenosis inhibitory agent selected from the group consisting of: (a) antiplatelet agents selected from the group consisting of (i) direct thrombin inhibitors and receptor antagonists, (ii) purinoceptor receptor antagonists, (iii) thromboxane inhibitors and receptor antagonists and (iv) platelet membrane glycoprotein receptor antagonists; (b) inhibitors of cell adhesion molecules, including (i) selectin inhibitors and (ii) integrin inhibitors; (c) anti-chemotactic agents; (d) interleukin receptor antagonists; and (e) intracellular signaling inhibitors selected from the group consisting of (i) protein kinase C inhibitors and protein tyrosine kinase inhibitors, (ii) modulators of intracellular protein tyrosine phosphatases, and (iii) inhibitors of src homology.sub.2 domains, the concentration of each agent within the solution being the concentration of that agent which is desired to be delivered locally to an operative vascular site in order to achieve a level of inhibitory effect at the operative vascular site and that is less than a concentration which would be required to provide the same level of inhibitory effect at the operative vascular site if the solution was applied systemically.

Claims Text - CLTX (63):
62. The solution of claim 51, wherein the restenosis inhibitory agents are selected from the group consisting of: antiplatelet agents including thrombin inhibitors and receptor antagonists, purinoceptor antagonists, thromboxane inhibitors and receptor antagonists and platelet membrane glycoprotein receptor antagonists; inhibitors of cell adhesion molecules, including selectin inhibitors and integrin inhibitors; anti-chemotactic agents; interleukin receptor antagonists; and intracellular signaling inhibitors including protein kinase C inhibitors and protein tyrosine kinase inhibitors, modulators of intracellular protein tyrosine phosphatases, inhibitors of src homology.sub.2 domains, and calcium channel antagonists.

Claims Text - CLTX (64):
63. The solution of claim 51, wherein the restenosis inhibitory agents are selected from the group consisting of: (a) antiplatelet agents selected from

the group consisting of (i) direct thrombin inhibitors and receptor antagonists, (ii) purinoceptor receptor antagonists, (iii) thromboxane inhibitors and receptor antagonists and (iv) platelet membrane glycoprotein receptor antagonists; (b) inhibitors of cell adhesion molecules, including (i) selectin inhibitors and (ii) integrin inhibitors; (c) anti-chemotactic agents; (d) interleukin receptor antagonists; and (e) intracellular signaling inhibitors selected from the group consisting of (i) protein kinase C inhibitors and protein tyrosine kinase **inhibitors, (ii) modulators of intracellular protein tyrosine phosphatases, and (iii) inhibitors of src** homology.sub.2 domains.

PGPUB-DOCUMENT-NUMBER: 20040127544

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040127544 A1

TITLE: Mannich base prodrugs of 3-(Pyrrol-2-yl-methylidene)-2-indolinone derivatives

PUBLICATION-DATE: July 1, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Moon, Malcolm Wilson	Kalamazoo	MI	US	
Morozowich, Walter	Kalamazoo	MI	US	
Gao, Ping	Portage	MI	US	
Tang, Peng Cho	Moraga	CA	US	

APPL-NO: 10/ 743909

DATE FILED: December 24, 2003

RELATED-US-APPL-DATA:

child 10743909 A1 20031224

parent continuation-of 09863804 20010524 US GRANTED

parent-patent 6710067 US

non-provisional-of-provisional 60207000 20000524 US

non-provisional-of-provisional 60225045 20000811 US

US-CL-CURRENT: 514/414, 548/465

ABSTRACT:

The present invention is directed to Mannich base prodrugs of certain 3-(pyrrol-2-ylmethylidene)-2-indolinone derivatives that modulate the activity of protein kinases ("PKs"). Pharmaceutical compositions comprising these compounds, methods of treating diseases related to abnormal PK activity utilizing pharmaceutical compositions comprising these compounds and methods of preparing them are also disclosed.

CROSS-REFERENCE

[0001] This application claims priority under 35 U.S.C. 119(e) to U.S. Provisional applications Serial No. 60/207,000 filed on May 24, 2000, and 60/225,045, filed on Aug. 11, 2000, the disclosures of which are incorporated herein by reference in their entirety.

---------- KWIC ---------

Detail Description Paragraph - DETX (687):
[0708] This assay is used to screen for **inhibitors of the tyrosine kinase Src**.

Detail Description Paragraph - DETX (734):
[0755] Vascular Permeability Assay

Detail Description Paragraph - DETX (735):
[0756] Increased vascular permeability in tumor-dependent angiogenesis is due to a loosening of gap junctions in response to vascular endothelial growth factor (VEGF). The Miles assay for vascular permeability (Miles and Miles, J. Physiol. 118: 228-257 (1952)) has been adapted to athymic mice in order to evaluate the ability of the compounds of the present invention to inhibit VEGF-induced vascular permeability in vivo.

PGPUB-DOCUMENT-NUMBER: 20040127542

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040127542 A1

TITLE:
1-(Pyrrolidin-1-ylmethyl)-3-(pyrrol-2-ylmethylidene)-2-in
dolinone derivatives

PUBLICATION-DATE: July 1, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Moon, Malcolm Wilson	Kalamazoo	MI	US	
Morozowich, Walter	Kalamazoo	MI	US	
Gao, Ping	Kalamazoo	MI	US	

APPL-NO: 10/ 429895

DATE FILED: May 5, 2003

RELATED-US-APPL-DATA:

child 10429895 A1 20030505

parent continuation-of 10243663 20020916 US ABANDONED

child 10243663 20020916 US

parent continuation-of 09863905 20010524 US GRANTED

parent-patent 6451838 US

non-provisional-of-provisional 60207000 20000524 US

non-provisional-of-provisional 60225045 20000811 US

US-CL-CURRENT: 514/414, 548/465

ABSTRACT:

The present invention is directed to 1-pyrrolidin-1-ylmethyl-3-(pyrrol-2-y-
lmethylidene)-2-indolinone derivatives that modulate the activity of protein
kinases ("PKs"). Pharmaceutical compositions comprising these compounds,
methods of treating diseases related to abnormal PK activity utilizing
pharmaceutical compositions comprising these compounds and methods of preparing
them are also disclosed.

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority under 35 U.S.C. 119(e) to U.S.
Provisional applications Serial Nos. 60/207,000 filed on May 24, 2000, and
60/225,045, filed on Aug. 11, 2000, the disclosures of which are incorporated
herein by reference in their entirety.

---------- KWIC ---------

Detail Description Paragraph - DETX (481):
[0674] This assay is used to screen for inhibitors of the tyrosine kinase Src.

Detail Description Paragraph - DETX (528):
[0721] Vascular Permeability Assay

Detail Description Paragraph - DETX (529):
[0722] Increased vascular permeability in tumor-dependent angiogenesis is due to a losening of gap junctions in response to vascular endothelial growth factor (VEGF). The Miles assay for vascular permeability (Miles and Miles, J. Physiol. 118: 228-257 (1952)) has been adapted to athymic mice in order to evaluate the ability of the compounds of the present invention to inhibit VEGF-induced vascular permeability in vivo.

PGPUB-DOCUMENT-NUMBER: 20040127453

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040127453 A1

TITLE: Method for treating diseases associated with abnormal kinase activity

PUBLICATION-DATE: July 1, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Lyons, John	Moraga	CA	US	
Rubinfeld, Joseph	Danville	CA	US	

APPL-NO: 10/ 206854

DATE FILED: July 26, 2002

RELATED-US-APPL-DATA:

child 10206854 A1 20020726

parent continuation-in-part-of 10071849 20020207 US PENDING

US-CL-CURRENT: 514/50

ABSTRACT:

Methods are provided for treating diseases associated with abnormal activity of kinases. The method comprises: administering a DNA methylation inhibitor to the patient in therapeutically effective amount; and administering a kinase inhibitor to the patient in therapeutically effective amount, such that the in vivo activity of the kinase is reduced relative to that prior to the treatment. The method can be used to treat cancer associated with abnormal activity of kinases such as phosphatidylinositol 3'-kinase (PI3K), protein kinases including serine/threonine kinases such as Raf kinases, protein kinase kinases such as MEK, and tyrosine kinases such as those in the epidermal growth factor receptor family (EGFR), platelet-derived growth factor receptor family (PDGFR), vascular endothelial growth factor receptor (VEGFR) family, nerve growth factor receptor family (NGFR), fibroblast growth factor receptor family (FGFR) insulin receptor family, ephrin receptor family, Met family, Ror family, c-kit family, Src family, Fes family, JAK family, Fak family, Btk family, Syk/ZAP-70 family, and Abl family.

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of U.S. patent application Ser. No. 10/071,849 entitled "Method for Treating Chronic Myelogenous Leukemia" filed on Feb. 7, 2002. The above application is incorporated herein by reference.

---------- KWIC ---------

Summary of Invention Paragraph - BSTX (63):
[0062] Examples of the inhibitors of the non-receptor tyrosine kinase from

the Src family include, but are not limited to, SU101 and CGP 57418B.

Detail Description Paragraph - DETX (67):
[0143] Vascular endothelial growth factor (VEGF) is a potent and specific angiogenic factor Verheul H M at al., The Role of Vascular Endothelial Growth Factor (VEGF) in Tumor Angiogenesis and Early Clinical Development of VEGF-Receptor Kinase Inhibitors. Clin Breast Cancer 2000 1 :S80-4. Originally identified for its ability to induce vascular permeability and stimulate endothelial cell growth, VEGF is now known to facilitate tumor growth. The family of VEGF receptors comprises high-affinity tyrosine kinase receptors VEGFR1, VEGFR2, and VEGFR3, of which VEGFR-Flk-1/KDR (VEGFR-2) is exclusively expressed in vascular endothelial cells.

Detail Description Paragraph - DETX (94):
[0170] The examples of Src inhibitors include, but are not limited to SU 101 and CGP 57418B. Broadbridge R J, et al., The Src homology-2 domains (SH2 domains) of the protein tyrosine kinase p56lck: structure, mechanism and drug design, Curr Drug Targets 2000 Dec; 1(4):365-86.

Claims Text - CLTX (28):
27. The method of claim 25, wherein the inhibitor of the Src family is selected from the group of SU101 and CGP 57418B.

PGPUB-DOCUMENT-NUMBER: 20040121362

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040121362 A1

TITLE: Identification and modulation of a G-protein coupled receptor (GPCR), RAI-3, associated with chronic obstructive pulmonary disease (COPD) and NF-kappaB and E-selectin regulation

PUBLICATION-DATE: June 24, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Whitney, Gena S.	Lawrenceville	NJ	US	
Opiteck, Gregory	Lawrenceville	NJ	US	
Garulacan, Leah Ann	Langhorne	PA	US	
Ramanathan, Chandra S.	Wallingford	CT	US	
McKinnon, Murray	Washington Crossing	PA	US	
Bennett, Kelley L.	Skillman	NJ	US	
Barber, Lauren E.	Higganum	CT	US	
Cacace, Angela	Clinton	CT	US	
Tsuchihashi, Zenta	Skillman	NJ	US	

APPL-NO: 10/ 600816

DATE FILED: June 20, 2003

RELATED-US-APPL-DATA:

non-provisional-of-provisional 60390850 20020620 US

non-provisional-of-provisional 60407006 20020829 US

US-CL-CURRENT: 435/6, 435/320.1 , 435/325 , 435/69.1 , 530/350 , 536/23.5

ABSTRACT:

The present invention describes a G-protein coupled receptor (GPCR) family member newly identified as being modified, e.g., phosphorylated, and associated with tyrosine phosphorylated activation complexes, following exposure of cells to smoke from tobacco burning substances, namely, cigarette smoke. This GPCR protein is RAI-3, which was first found to be phosphorylated in cells treated with cigarette smoke and to be associated with other proteins activated in cigarette smoke treated cells by virtue of the present invention. Because cigarette smoke is considered to be a major causative factor of chronic obstructive pulmonary disease (COPD) and disorders and conditions related thereto, the RAI-3 protein is newly provided as a cellular drug target for screening, discovering, and identifying modulators for the treatment and/or prevention of COPD and its related disorders and conditions, such as emphysema and chronic bronchitis. In accordance with the present invention RAI-3 modulators, e.g., agonists and antagonists, can be used as therapeutics in the treatment of COPD and numerous other diseases and disorders that are associated with regulation of NF-.kappa.B and/or its associated or interacting signaling molecules. This invention further provides SNPs of RAI-3, e.g., for determining COPD association in individuals.

[0001] This application claims benefit to provisional application U.S. Serial No. 60/390,850 filed Jun. 20, 2002; and to provisional application U.S. Serial No. 60/407,006, filed Aug. 29, 2002, under 35 U.S.C. 119(e). The entire teachings of the referenced applications are incorporated herein by reference.

---------- KWIC --------

Brief Description of Drawings Paragraph - DRTX (40):

[0106] This strategy was employed based on the following nonlimiting hypothesis according to this invention: Exposure to smoke induces oxidative stress, inhibits phosphatases, activates Src and transactivates EGFR, and potentially other receptor tyrosine kinases. This activation contributes to the transcription of mucin and cytokine genes which, in turn, contribute to the cause and symptoms of COPD. Compounds that are able to inhibit this activation, e.g., by affecting cellular proteins and/or peptides that are induced, activated and/or modified following cigarette smoke exposure, are thus reasoned to be useful as drugs for treating COPD.

Brief Description of Drawings Paragraph - DRTX (315):

[0370] The central role of NF-.kappa.B activation in bronchiolar epithelium in coordinating airway inflammation has been demonstrated in a number of mouse and cellular models where chemokine, eotaxin, IL-8, MMP-9, and iNOS synthase expression are enhanced leading to neutrophil and eosinophil infiltration and tissue damage (M. E. Poynter, 2002, Am. J. Pathol., 160:1325-1334; D-W. Jeong, 2002, J. Biol. Chem., 277:17871-17876; A. Hozumi et al., 2001, Am. J. Physiol. Lung Cell Mol. Physiol., 281:L1444-L1452; R. S. Smith et al., 2001, J. Immunol., 167:366-374; R. W. Ganster et al., 2001, Proc. Natl. Acad. Sci. USA, 98:8638-8643; and H. Takizawa, 1999, J. Immunol., 162:4705-4711). In addition, NF-.kappa.B has been shown to regulate aquaporin 5, a major water channel that is expressed in alveolar, tracheal and upper bronchial epithelium, thereby contributing not only to lung inflammation, but also to airway edema (J. E. Towne et al., 2001, J. Biol. Chem., 276:18657-18664). Mucin production by specialized epithelial cells has also been shown to be regulated by NF-.kappa.B (Seuningen et al., 2001. Front. Biosci., 6:D1216-D1234; and J.-D. Li et al., 1998, Proc. Natl. Acad. Sci. USA, 95:5718-5723). Thus, using antagonist or agonist modulators of RAI-3 that target lung epithelial cells offers a novel utility as therapeutics for many diseases of the lung.

PGPUB-DOCUMENT-NUMBER: 20040106615

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040106615 A1

TITLE: Protein kinase inhibitors and uses thereof

PUBLICATION-DATE: June 3, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Cochran, John	Marshfield	MA	US	
Green, Jeremy	Burlington	MA	US	
Hale, Michael R.	Bedford	MA	US	
Ledford, Brian	Attleboro	MA	US	
Maltais, Francois	Tewksbury	MA	US	
Nanthakumar, Suganthini	Newton	MA	US	

APPL-NO: 10/ 639784

DATE FILED: August 12, 2003

RELATED-US-APPL-DATA:

non-provisional-of-provisional 60403256 20020814 US

non-provisional-of-provisional 60416802 20021008 US

US-CL-CURRENT: 514/242, 514/247 , 514/252.03 , 514/275 , 544/183 , 544/238 , 544/331

ABSTRACT:

Described herein are compounds that are useful as protein kinase inhibitors having the formulae I and V: 1
or a pharmaceutically acceptable salt thereof, wherein Ring B, Z.sup.1, Z.sup.2, U, T, m, n, p, Q, Q', R.sup.1, R.sup.2, R.sup.x, R.sup.3, and R.sup.6 are as defined herein. These compounds, and pharmaceutically acceptable compositions thereof, are useful for treating or lessening the severity of a variety of disorders, including stroke, inflammatory disorders, autoimmune diseases such as SLE lupus and psoriasis, proliferative disorders such as cancer, and conditions associated with organ transplantation.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority to U.S. Provisional Patent Applications 60/403,256 filed Aug. 14, 2002 and 60/416,802 filed Oct. 8, 2002, the contents of which are incorporated herein by reference.

---------- KWIC ---------

Summary of Invention Paragraph - BSTX (77):
[0076] Based on published studies, Src kinases are considered as potential therapeutic targets for various human diseases. Mice that are deficient in Src develop osteopetrosis, or bone build-up, because of depressed bone resorption by osteoclasts. This shows that osteoporosis resulting from abnormally high

bone resorption is treated by inhibiting Src. Soriano et al., Cell 1992, 69, 551 and Soriano et al., Cell 1991, 64, 693.

Summary of Invention Paragraph - BSTX (78):
[0077] Suppression of arthritic bone destruction has been achieved by the overexpression of CSK in rheumatoid synoviocytes and osteoclasts. Takayanagi et al., J. Clin. Invest. 1999, 104, 137. CSK, or C-terminal Src kinase, phosphorylates and thereby inhibits Src catalytic activity. This implies that Src inhibition may prevent joint destruction that is characteristic in patients suffering from rheumatoid arthritis. Boschelli et al., Drugs of the Future 2000, 25(7), 717.

Summary of Invention Paragraph - BSTX (80):
[0079] A number of studies have linked Src expression to cancers such as colon, breast, hepatic and pancreatic cancer, certain B-cell leukemias and lymphomas. Talamonti et al., J. Clin. Invest. 1993, 91, 53; Lutz et al., Biochem. Biophys. Res. 1998 243, 503; Rosen et al., J. Biol. Chem. 1986, 261, 13754; Bolen et al., Proc. Natl. Acad. Sci. USA 1987, 84, 2251; Masaki et al., Hepatology 1998, 27, 1257; Biscardi et al., Adv. Cancer Res. 1999, 76, 61; Lynch et al., Leukemia 1993, 7, 1416. Furthermore, antisense Src expressed in ovarian and colon tumor cells has been shown to inhibit tumor growth. Wiener et al., Clin. Cancer Res., 1999, 5, 2164; Staley et al., Cell Growth Diff. 1997, 8, 269.

Summary of Invention Paragraph - BSTX (99):
[0097] It has now been found that compounds of this invention, and compositions thereof, are effective as protein kinase inhibitors. In certain embodiments, the present compounds are inhibitors of ERK2, AKT3, GSK3, p70s6k, PDK1, Aurora-2, ROCK, SRC, SYK, ZAP70, JNK3, JAK3, TEC, LCK, FLT3, and/or CDK2. These compounds have the general formulae I and V: 2

Summary of Invention Paragraph - BSTX (361):
[0358] The activity of a compound utilized in this invention as an inhibitor of ERK2, AKT3, GSK3, p70s6k, PDK1, Aurora-2, ROCK, SRC, SYK, ZAP70, JNK3, JAK3, TEC, LCK, FLT3, and/or CDK2, may be assayed in vitro, in vivo or in a cell line. In vitro assays include assays that determine inhibition of either the phosphorylation activity or ATPase activity of activated ERK2, AKT3, GSK3, p70s6k, PDK1, Aurora-2, ROCK, SRC, SYK, ZAP70, JNK3, JAK3, TEC, LCK, FLT3, and/or CDK2. Alternate in vitro assays quantitate the ability of the inhibitor to bind to ERK2, AKT3, GSK3, p70s6k, PDK1, Aurora-2, ROCK, SRC, SYK, ZAP70, JNK3, JAK3, TEC, LCK, FLT3, and/or CDK2. Inhibitor binding may be measured by radiolabelling the inhibitor prior to binding, isolating the inhibitor/ERK2, inhibitor/AKT3, inhibitor/GSK3, inhibitor/p70s6k, inhibitor/PDK1, inhibitor/Aurora-2, inhibitor/ROCK, inhibitor/SRC, inhibitor/SYK, inhibitor/ZAP70, inhibitor/JNK3, inhibitor/JAK3, inhibitor/TEC, inhibitor/LCK, inhibitor/FLTS, or inhibitor/CDK2, complex and determining the amount of radiolabel bound. Alternatively, inhibitor binding may be determined by running a competition experiment where new inhibitors are incubated with ERK2, AKT3, GSK3, p70s6k, PDK1, Aurora-2, ROCK, SRC, SYK, ZAP70, JNK3, JAK3, TEC, LCK, FLT3, and/or CDK2 bound to known radioligands. Detailed conditions for assaying a compound utilized in this invention as an inhibitor of ERK2, AKT3, GSK3, p70s6k, PDK1, Aurora-2, ROCK, SRC, SYK, ZAP70, JNK3, JAK3, TEC, LCK, FLT3, and CDK2 kinase are set forth in the Examples below.

Summary of Invention Paragraph - BSTX (369):
[0366] As used herein, the term "inhibitorily active metabolite or residue thereof" means that a metabolite or residue thereof is also an inhibitor of ERK2, AKT3, GSK3, p70s6k, PDK1, Aurora-2, ROCK, SRC, SYK, ZAP70, JNK3, JAK3, TEC, LCK, FLT3, and/or CDK2 kinase.

Summary of Invention Paragraph - BSTX (385):
[0382] According to another embodiment, the invention relates to a method of inhibiting ERK2, AKT3, GSK3, p70s6k, PDK1, Aurora-2, ROCK, SRC, SYK, ZAP70, JNK3, JAK3, TEC, LCK, FLT3, and/or CDK2 kinase activity in a biological sample comprising the step of contacting said biological sample with a compound of this invention, or a composition comprising said compound.

Summary of Invention Paragraph - BSTX (389):
[0386] According to another embodiment, the invention relates to a method of inhibiting ERK2, AKT3, GSK3, p70s6k, PDK1, Aurora-2, ROCK, SRC, SYK, ZAP70, JNK3, JAK3, TEC, LCK, FLT3, and/or CDK2 kinase activity in a patient comprising the step of administering to said patient a compound of the present invention, or a composition comprising said compound.

Summary of Invention Paragraph - BSTX (414):
[0411] The terms "SRC-mediated disease" or "SRC-mediated condition", as used herein mean any disease or other deleterious condition in which SRC is known to play a role. The terms "SRC-mediated disease" or "SRC-mediated condition" also mean those diseases or conditions that are alleviated by treatment with a SRC inhibitor. Accordingly, another embodiment of the present invention relates to treating or lessening the severity of one or more diseases in which SRC is known to play a role. Specifically, the present invention relates to a method of treating or lessening the severity of a disease or condition selected from hypercalcemia, osteoporosis, osteoarthritis, cancer, symptomatic treatment of bone metastasis, and Paget's disease, wherein said method comprises administering to a patient in need thereof a composition according to the present invention.

Summary of Invention Paragraph - BSTX (437):
[0434] In addition, compounds of the present invention may be capable of inhibiting the expression of inducible pro-inflammatory proteins. Therefore, other "JNK-mediated diseases" or "conditions" which may be treated by the compounds of this invention include edema, analgesia, fever and pain, such as neuromuscular pain, headache, cancer pain, dental pain and arthritis pain.

Detail Description Paragraph - DETX (147):
SRC Inhibition Assay:

Detail Description Paragraph - DETX (148):
[0540] The compounds of the present invention were evaluated as inhibitors of human Src kinase using either a radioactivity-based assay or spectrophotometric assay.

Detail Description Paragraph - DETX (149):
[0541] Src Inhibition Assay A: Radioactivity-Based Assay

Detail Description Paragraph - DETX (150):
[0542] The compounds of the present invention were assayed as inhibitors of full length recombinant human Src kinase (from Upstate Biotechnology, Cat. No. 14-117) expressed and purified from baculo viral cells. Src kinase activity was monitored by following the incorporation of .sup.33P from ATP into the tyrosine of a random poly Glu-Tyr polymer substrate of composition, Glu:Tyr=4:1 (Sigma, Cat. No. P-0275). The final concentrations of the assay components were: 0.05 M HEPES (pH 7.6), 10 mM MgCl.sub.2, 2 mM DTT, 0.25 mg/ml BSA, 10 .mu.M ATP (1-2 .mu.Ci .sup.33P-ATP per reaction), 5 mg/ml poly Glu-Tyr, and 1-2 units of recombinant human Src kinase. In a typical assay, all the reaction components with the exception of ATP were pre-mixed and aliquoted into assay plate wells. Compounds of the present invention were dissolved in DMSO and

added to the wells to give a final DMSO concentration of 2.5%. The assay plate was incubated at 30.degree. C. for 10 min before initiating the reaction with .sup.33P-ATP. After 20 min of reaction, the reactions were quenched with 150 .mu.l of 10% trichloroacetic acid (TCA) containing 20 mM Na.sub.3PO.sub.4. The quenched samples were then transferred to a 96-well filter plate (Whatman, UNI-Filter GF/F Glass Fiber Filter, Cat No. 7700-3310) installed on a filter plate vacuum manifold. Filter plates were washed four times with 10% TCA containing 20 mM Na.sub.3PO.sub.4 and then 4 times with methanol. 200 .mu.l of scintillation fluid was then added to each well. The plates were sealed and the amount of radioactivity associated with the filters was quantified on a TopCount scintillation counter. The radioactivity incorporated was plotted as a function of the compound of the present invention concentration. The data was fitted to a competitive inhibition kinetics model to give the K.sub.i values for the compounds of the present invention.

Detail Description Paragraph - DETX (151):
[0543] Src Inhibition Assay B: Spectrophotometric Assay

Detail Description Paragraph - DETX (155):
[0547] Compounds of the present invention were found to be inhibitors of SRC.

PGPUB-DOCUMENT-NUMBER: 20040097531

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040097531 A1

TITLE: Inhibitors of c-Jun N-terminal kinases (JNK) and other protein kinases

PUBLICATION-DATE: May 20, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Ledeboer, Mark	Acton	MA	US	
Wang, Jian	Boston	MA	US	
Moon, Young Choon	Belle Mead	NJ	US	

APPL-NO: 10/ 616560

DATE FILED: July 9, 2003

RELATED-US-APPL-DATA:

non-provisional-of-provisional 60395202 20020709 US

US-CL-CURRENT: 514/275, 514/341 , 514/342 , 544/331 , 546/270.4 , 546/271.4 , 546/272.7

ABSTRACT:

The present invention provides compounds of formula I: 1 or a pharmaceutically acceptable derivative thereof, wherein R.sup.1, R.sup.2, A, G, and W are as described in the specification. These compounds are inhibitors of protein kinase, particularly inhibitors of JNK, a mammalian protein kinase involved cell proliferation, cell death and response to extracellular stimuli, Lck, Src, and Aurora kinases. The invention also relates to methods for producing these inhibitors. The invention also provides pharmaceutical compositions comprising the inhibitors of the invention and methods of utilizing those compositions in the treatment and prevention of various disorders.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority to U.S. Provisional Patent Application 60/395,202, filed Jul. 9, 2002, which is hereby incorporated by reference in its entirety.

---------- KWIC --------

Summary of Invention Paragraph - BSTX (2):

[0002] The present invention relates to **inhibitors of protein kinase, especially c-Jun N-terminal kinases (JNK) the Src**-family of kinases, including Lck, which are members of the mitogen-activated protein (MAP) kinase family, and the Aurora family, including Aurora-2, which are serine/threonine kinases. JNK, Src, Lck, and Aurora-2 have been implicated in a number of different human diseases. The invention also provides pharmaceutical compositions comprising the **inhibitors of the invention and methods of utilizing those compositions in**

the treatment and prevention of various disorders in which JNK, Src, Lck, and/or Aurora-2 kinases play a role.

Summary of Invention Paragraph - BSTX (17):
[0016] Based on published studies, Src kinases are considered as potential therapeutic targets for various human diseases. Mice that are deficient in Src develop osteopetrosis, or bone build-up, because of depressed bone resorption by osteoclasts. This suggests that osteoporosis resulting from abnormally high bone resorption can be treated by inhibiting Src [Soriano et al., Cell, 69:551 (1992) and Soriano et al., Cell, 64: 693 (1991)].

Summary of Invention Paragraph - BSTX (18):
[0017] Suppression of arthritic bone destruction has been achieved by the overexpression of CSK in rheumatoid synoviocytes and osteoclasts [Takayanagi et al., J. Clin. Invest., 104:137 (1999)]. CSK, or C-terminal Src kinase, phosphorylates and thereby inhibits Src catalytic activity. This implies that Src inhibition may prevent joint destruction that is characteristic in patients suffering from rheumatoid arthritis [Boschelli et al., Drugs of the Future 2000, 25(7):717 (2000)].

Summary of Invention Paragraph - BSTX (20):
[0019] A number of studies have linked Src expression to cancers such as colon, breast, hepatic and pancreatic cancer, certain B-cell leukemias and lymphomas [Talamonti et al., J. Clin. Invest., 91:53 (1993); Lutz et al., Biochem. Biophys. Res. 243:503 (1998); Rosen et al., J. Biol. Chem., 261:13754 (1986); Bolen et al., Proc. Natl. Acad. Sci. USA, 84:2251 (1987); Masaki et al., Hepatology, 27:1257 (1998); Biscardi et al., Adv. Cancer Res., 76:61 (1999); Lynch et al., Leukemia, 7:1416 (1993)]. Furthermore, antisense Src expressed in ovarian and colon tumor cells has been shown to inhibit tumor growth [Wiener et al., Clin. Cancer Res., 5:2164 (1999) and Staley et al., Cell Growth Diff., 8:269 (1997)].

Summary of Invention Paragraph - BSTX (31):
[0030] There is a continued need to develop potent inhibitors of JNKs, Src family kinases, and Aurora family kinases that are useful in treating or preventing various conditions associated with JNK, Src, and Aurora activation.

Summary of Invention Paragraph - BSTX (36):
[0034] The compounds and pharmaceutical compositions of the present invention are useful as inhibitors of c-Jun N-terminal kinases (JNK) Src family kinases, including Src and Lck, and Aurora family kinases, including Aurora-2. Thus, they are also useful in methods for treating or preventing a variety of disorders, such as heart disease, immunodeficiency disorders, inflammatory diseases, allergic diseases, autoimmune diseases, destructive bone disorders such as osteoporosis, proliferative disorders, infectious diseases and viral diseases. The compositions are also useful in methods for preventing cell death and hyperplasia and therefore may be used to treat or prevent reperfusion/ischemia in stroke, heart attacks, and organ hypoxia. The compositions are also useful in methods for preventing thrombin-induced platelet aggregation. The compositions are especially useful for disorders such as chronic myelogenous leukemia (CML), rheumatoid arthritis, asthma, osteoarthritis, ischemia, cancer, liver disease including hepatic ischemia, heart disease such as myocardial infarction and congestive heart failure, pathologic immune conditions involving T cell activation and neurodegenerative disorders.

Summary of Invention Paragraph - BSTX (165):
[0162] According to another embodiment, the invention provides a method of inhibiting JNK, Src, Lck, or Aurora-2kinase activity in a biological sample.

This method comprises the step of contacting said biological sample with a compound of formula I. According to a preferred embodiment, the invention relates to a method of inhibiting JNK, Src, Lck, or Aurora-2 kinase activity in a biological sample comprising the step of contacting said biological sample with a compound of formula IIa, IIb, IVa, or IVb.

Summary of Invention Paragraph - BSTX (167):
[0164] Inhibition of JNK, Src, Lck, or Aurora-2 kinase activity in a biological sample is useful for a variety of purposes which are known to one of skill in the art. Examples of such purposes include, but are not limited to, blood transfusion, organ-translplantation, biological specimen storage, and biological assays.

Summary of Invention Paragraph - BSTX (168):
[0165] Compounds of formula I or derivatives (e.g., salts) thereof may be formulated into compositions. In a preferred embodiment, the composition is a pharmaceutically acceptable composition. In one embodiment, the composition comprises an amount of a compound effective to inhibit a protein kinase, particularly JNK, Src, Lck, or Aurora-2, in a biological sample or in a patient. In another embodiment, compounds of this invention and pharmaceutical compositions thereof, which comprise an amount of the compound effective to treat or prevent an JNK, Src, Lck, or Aurora-2-mediated condition and a pharmaceutically acceptable carrier, adjuvant, or vehicle, may be formulated for administration to a patient.

Summary of Invention Paragraph - BSTX (169):
[0166] The amount effective to inhibit protein kinase, for example, JNK, Src, Lck, or Aurora-2, is one that measurably inhibits the kinase activity where compared to the activity of the enzyme in the absence of an inhibitor. "Measurable inhibition" means a measurable change in activity between a sample containing said inhibitor and a sample containing said protein kinase only. Any method may be used to determine inhibition, such as, for example, the biological testing examples described below.

Summary of Invention Paragraph - BSTX (190):
[0187] The compounds of this invention are inhibitors of JNK, Src, Lck, or Aurora-2 kinase as determined by enzymatic assay. Accordingly, these compounds are useful for treating JNK-, Src-, Lck-, or Aurora-2-mediated diseases or conditions.

Summary of Invention Paragraph - BSTX (193):
[0190] The activity of the compounds of this invention as kinase inhibitors may be assayed in vitro, in vivo or in a cell line. In vitro assays include assays that determine inhibition of either the kinase activity or ATPase activity of activated enzyme, for example JNK, Lck, Src or Aurora-2. Alternate in vitro assays quantitate the ability of the inhibitor to bind to JNK, Lck, Src, or Aurora-2 and may be measured either by radiolabelling the inhibitor prior to binding, isolating the inhibitor/JNK, inhibitor/Lck, or inhibitor/Src complex and determining the amount of radiolabel bound, or by running a competition experiment where new compounds are incubated with JNK, Lck, Src, or Aurora-2 bound to known radioligands. One may use any type or isoform of JNK, Lck, Src, or Aurora-2, depending upon which JNK, Lck, Src, or Aurora-2 type or isoform is to be inhibited. The details of the conditions used for the enzymatic assays are set forth in the Examples hereinbelow.

Summary of Invention Paragraph - BSTX (203):
[0200] In addition, JNK inhibitors of the present invention may be capable of inhibiting the expression of inducible pro-inflammatory proteins. Therefore, other "JNK-mediated diseases", "disorder" or "conditions" which may

be treated by the compounds of this invention include edema, analgesia, fever and pain such as neuromuscular pain, headache, cancer pain, dental pain and arthritis pain.

Summary of Invention Paragraph - BSTX (204):
[0201] The compounds of this invention are also useful as inhibitors of Src-family kinases, especially Src. For a general review of these kinases see Thomas and Brugge, Annu. Rev. Cell Dev. Biol. 13:513 (1997); Lawrence and Niu, Pharmacol. Ther. 77:81 (1998); and Tatosyan and Mizenina, Biochemistry (Moscow) 65:49 (2000). Accordingly, these compounds are useful for treating Src-mediated diseases, disorders or conditions.

Summary of Invention Paragraph - BSTX (218):
[0215] Each of the aforementioned methods directed to the inhibition of JNK, Lck, Src, or Aurora-2, or the treatment of a disease alleviated thereby, is preferably carried out with a preferred compound of formula I, IIa, IIb, IVa, or IVb, as described above. More preferably, each of the aforementioned methods is carried out with a preferred compound of formula IIa, IIb, IVa, or IVb.

Detail Description Paragraph - DETX (53):
[0255] The following examples demonstrate how the compounds of this invention may be tested as inhibitors of c-Jun-N-terminal, Src, and Lck kinases.

Detail Description Paragraph - DETX (69):
[0264] The compounds of this invention can be evaluated as inhibitors of human Src kinase using either a radioactivity-based assay or spectrophotometric assay.

Detail Description Paragraph - DETX (70):
Src Inhibition Assay A: Radioactivity-Based Assay

Detail Description Paragraph - DETX (71):
[0265] The compounds can be assayed as inhibitors of full length recombinant human Src kinase (from Upstate Biotechnology, cat. no. 14-117) expressed and purified from baculo viral cells. Src kinase activity is monitored by following the incorporation of .sup.33P from ATP into the tyrosine of a random poly Glu-Tyr polymer substrate of composition, Glu:Tyr=4:1 (Sigma, cat. no. P-0275). The following are the final concentrations of the assay components: 0.05 M HEPES, pH 7.6, 10 mM MgCl.sub.2, 2 mM DTT, 0.25 mg/ml BSA, 10 .mu.M ATP (1-2 .mu.Ci .sup.33P-ATP per reaction), 5 mg/ml poly Glu-Tyr, and 1-2 units of recombinant human Src kinase. In a typical assay, all the reaction components with the exception of ATP are pre-mixed and aliquoted into assay plate wells. Inhibitors dissolved in DMSO are added to the wells to give a final DMSO concentration of 2.5%. The assay plate is incubated at 30.degree. C. for 10 min before initiating the reaction with .sup.33P-ATP. After 20 min of reaction, the reactions are quenched with 150 .mu.l of 10% trichloroacetic acid (TCA) containing 20 mM Na.sub.3PO.sub.4. The quenched samples are then transferred to a 96-well filter plate (Whatman, UNI-Filter GF/F Glass Fiber Filter, cat no. 7700-3310) installed on a filter plate vacuum manifold. Filter plates are washed four times with 10% TCA containing 20 mM Na.sub.3PO.sub.4 and then 4 times with methanol. 200 .mu.l of scintillation fluid is then added to each well. The plates are sealed and the amount of radioactivity associated with the filters is quantified on a TopCount scintillation counter. The radioactivity incorporated is plotted as a function of the inhibitor concentration. The data is fitted to a competitive inhibition kinetics model to get the K.sub.i for the compound.

Detail Description Paragraph - DETX (72):
Src Inhibition Assay B: Spectrophotometric Assay

Claims Text - CLTX (21):
21. A method of inhibiting JNK, Lck, Src, or Aurora-2 kinase activity in a biological sample comprising the step of contacting said biological sample with: (a) a compound according to claim 1; or (b) a composition according to claim 19.

PGPUB-DOCUMENT-NUMBER: 20040082664

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040082664 A1

TITLE: Derivatives of hydroxyphenyl, a method for preparing thereof and their pharmaceutical composition

PUBLICATION-DATE: April 29, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Won, Jongwha	Seongnam-si		KR	
Lee, Keunhyeung	Seoul		KR	
Park, Seehyoung	Suwon-si		KR	
Kim, Sung-Joo	Yongin-si		KR	
Yun, Su-Young	Seongnam-si		KR	
Kang, Mi-Ae	Gunpo-si		KR	
Hur, Yun-Gyoung	Hwaseong-gun		KR	
Youn, Jeehee	Seoul		KR	
Yun, Yungdae	Seoul		KR	
Park, Doohong	Seoul		KR	
Oh, Jaetaek	Seoul		KR	

APPL-NO: 10/ 411772

DATE FILED: April 11, 2003

FOREIGN-APPL-PRIORITY-DATA:

COUNTRY	APPL-NO	DOC-ID	APPL-DATE
KR	KR/2002-0020481	2002KR-KR/2002-0020481	April 15, 2002

US-CL-CURRENT: 514/649

ABSTRACT:

The present invention relates to derivatives of hydroxyphenyl, a method for preparing thereof and their pharmaceutical composition, more particularly the compounds of the present invention specifically inhibit the activation of T lymphocyte by src homology region 2(SH2) domain of T lymphocyte (lck), so that they can be used for the treatment, prevention and/or diagnosis of graft rejection, autoimmune diseases, inflammatory diseases, etc.

---------- KWIC --------

Abstract Paragraph - ABTX (1):

The present invention relates to derivatives of hydroxyphenyl, a method for preparing thereof and their pharmaceutical composition, more particularly the compounds of the present invention specifically inhibit the activation of T lymphocyte by src homology region 2(SH2) domain of T lymphocyte (lck), so that they can be used for the treatment, prevention and/or diagnosis of graft rejection, autoimmune diseases, inflammatory diseases, etc.

Summary of Invention Paragraph - BSTX (107):

[0104] The compounds of the present invention inhibit in vitro binding of lck SH2 domain to specific peptide ligand. More particularly, the compounds of

the present invention selectively bind the lck SH2 domain and interfere with the formation or stabilization of signaling complexes formed by proteins containing one or more SH2 domains and their natural ligands. Therefore, the compounds can be used to treat or prevent the diseases mediated by such complexes. Like this, the compounds of the present invention can be used for inhibiting the SH2-mediated cellular functions of Src based protein tyrosine kinases. The Src based protein tyrosine kinase comprises Src, Fyn, Yes, Lck, Lyn and Blk.

Detail Description Paragraph - DETX (304):

[0280] From the 3.sup.rd week after primary immunization, the extent of edema and the joint swelling was monitored everyday to determine arthritic index. Arthritic score was determined based on the criteria listed in Table 4. Arthitic index is the sum of the arthritic scores of all 4 legs and, therefore, maximum arthritis index of 16 can be achieved (e.g., [4 (maximum arthritic score)/leg].times.[4 legs/mouse]=16 (maximum arthritis index/mouse)). The of each compound to inhibit collagen-induced tis was presented by the mean arthritis index.+-.standard deviation of each mouse group. The results were presented in Table 5.

Detail Description Paragraph - DETX (312):

[0286] As explained hereinbefore, the compounds of the present invention inhibited the molecular interactions of lckSH2 and its cognate peptide PYEEI and TCR-induced IL-2 gene expression, resulting in immunosuppression in vitro and in vivo. Therefore, the compounds of the invention can be effectively used for inhibiting lck SH2 domain or Src based protein tyrosine kinase SH2 domain and suppressing allograft rejection, autoimmune diseases and inflammatory diseases. Also, the compounds of the present invention have sufficiently high activity at low dosages and low side effects being observed in currently used therapeutics for arthritis, so that they can be used for the treatment or prevention of rheumatoid arthritis or inflammatory diseases.

Claims Text - CLTX (5):

4. A pharmaceutical composition for use in inhibiting activation of src homology region 2 domain of T lymphocyte cell kinase comprising the derivatives or their pharmaceutically acceptable salts of claim 1 as an effective ingredient.

PGPUB-DOCUMENT-NUMBER: 20040063712

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040063712 A1

TITLE: Pyrrolopyridazine compounds and methods of use thereof for the treatment of proliferative disorders

PUBLICATION-DATE: April 1, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Salvati, Mark	Lawrenceville	NJ	US	
Barbosa, Stephanie A.	Lambertville	NJ	US	
Chen, Zhong	Cranbury	NJ	US	
Hunt, John T.	Princeton	NJ	US	

APPL-NO: 10/ 396197

DATE FILED: March 25, 2003

RELATED-US-APPL-DATA:

non-provisional-of-provisional 60368249 20020328 US

non-provisional-of-provisional 60402118 20020808 US

US-CL-CURRENT: 514/248, 424/143.1 , 424/145.1 , 514/109 , 514/217.03 , 514/251 , 514/263.31 , 514/263.4 , 514/269 , 514/283 , 514/410 , 514/449 , 514/45 , 514/49 , 544/235

ABSTRACT:

Disclosed are pyrrolopyridazine compounds, methods of preparing such compounds, and their use for the treatment of proliferative, inflammatory, and other disorders.

RELATED APPLICATIONS

[0001] This application claims priority benefit under Title 35 .sctn.119(e) of U.S. provisional Application No. 60/368,249, filed Mar. 28, 2002, and No. 60/402,118, filed Aug. 8, 2002, the contents of which are herein incorporated by reference.

---------- KWIC ---------

Summary of Invention Paragraph - BSTX (10):

[0009] The Src family of cytoplasmic protein tyrosine kinases consists of at least eight members (Src, Fyn, Lyn, Yes, Lck, Fgr, Hck and Blk) that participate in a variety of signaling pathways [Schwartzberg, P. L., Oncogene, 17, 1463 (1998)]. The prototypical member of this tyrosine kinase family is p60.sup.src (Src). Src is involved in proliferation and migration responses in many cell types. In limited studies, Src activity has been shown to be elevated in breast, colon (.about.90%), pancreatic (>90%) and liver (>90%) tumors. Greatly increased Src activity is also associated with metastasis (>90%) and poor prognosis. Antisense Src message impedes growth

of colon tumor cells in nude mice [Staley et al., Cell Growth & Differentation, 8, 269 (1997)], suggesting that Src inhibitors should slow tumor growth. In addition to its role in cell proliferation, Src also acts in stress response pathways, including the hypoxia response. Previous studies have shown that colonic tumor cells genetically engineered to express antisense Src message form tumors demonstrating reduced vascularization in nude mouse models [Ellis, et al., J. Biol. Chem., 273, 1052 (1998)], suggesting that Src inhibitors would be anti-angiogenic as well as anti-proliferative.

Summary of Invention Paragraph - BSTX (11):

[0010] Apart from its role in cancer, Src also appears to play a role in osteoporosis. Mice genetically engineered to be deficient in src production were found to exhibit osteopetrosis, the failure to resorb bone [Soriano, P., Cell, 64, 693 (1991); Boyce, B. F., J. Clin. Invest., 90, 1622 (1992)]. This defect was characterized by a lack of osteoclast activity. Since osteoclasts normally express high levels of Src, inhibition of Src kinase activity may be useful in the treatment of osteoporosis [Missbach, M., Bone, 24, 437 (1999)].

Detail Description Paragraph - DETX (103):

[0134] It has been discovered that pyrrolopyridazines of the invention are inhibitors of protein kinases. More specifically, certain pyrrolopyridazines inhibit the effects of receptor tyrosine kinases and serine/threonine kinases, a property of value in the treatment of disease states associated with hyperproliferation, angiogenesis, increased vascular permeability, and inflammation, such as cancer and inflammatory disease. In particular, the compounds of formula I and their salts, solvates, and stereoisomers are expected to inhibit the growth of primary and recurrent solid tumors by antiproliferative and/or antiangiogenic mechanisms. The solid tumors include, for example, cancers of the bladder, squamous cell, head, colorectal, oesophageal, gynecological (such as ovarian), pancreas, breast, prostate, lung, vulva, skin, brain, genitourinary tract, lymphatic system (such as thyroid), stomach, larynx and lung

Detail Description Paragraph - DETX (132):

[0163] In addition, protein kinase inhibitors of this invention also exhibit inhibition of the expression of inducible pro-inflammatory proteins such as prostaglandin endoperoxide synthase-2 (PGHS-2), also referred to as cyclooxygenase-2 (COX-2). Accordingly, additional conditions which may be treated or prevented by appropriate administration of compounds of the invention include edema, analgesia, fever and pain, such as neuromuscular pain, headache, pain caused by cancer, dental pain and arthritis pain.

Detail Description Paragraph - DETX (133):

[0164] In the field of medical oncology, it is normal practice to combine different agents for treatment of patients with cancer. Thus, a compound of formula I may optionally be combined with other components, such as antiproliferative, antiangiogenic and/or vascular permeability reducing agents. Additionally, surgery, radiotherapy or chemotherapy may optionally be utilized in conjunction with administration of compounds of formula I. Accordingly, the compound of formula I may be administered alone or combined with the administration of one or more other therapeutic agents, substances and/or treatments.

PGPUB-DOCUMENT-NUMBER: 20040023963

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040023963 A1

TITLE: Inhibitors of c-Jun N-terminal kinases (JNK) and other protein kinases

PUBLICATION-DATE: February 5, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Cao, Jingrong	Newton	MA	US	
Green, Jeremy	Burlington	MA	US	
Moon, Young-Choon	Lexington	MA	US	
Wang, Jian	Boston	MA	US	
Ledeboer, Mark	Acton	MA	US	
Harrington, Edmund	South Boston	MA	US	
Gao, Huai	Natick	MA	US	

APPL-NO: 10/ 437666

DATE FILED: May 14, 2003

RELATED-US-APPL-DATA:

child 10437666 A1 20030514

parent division-of 10121035 20020410 US PENDING

non-provisional-of-provisional 60283621 20010413 US

non-provisional-of-provisional 60329440 20011015 US

non-provisional-of-provisional 60292974 20010523 US

US-CL-CURRENT: 514/242, 514/227.8 , 514/235.8 , 514/252.01 , 514/275 , 544/112 , 544/182 , 544/238 , 544/331 , 544/60

ABSTRACT:

The present invention provides compounds of formula I: 1
or a pharmaceutically acceptable derivative o thereof, wherein A, B, R.sup.a, R.sup.1, R.sup.2, R.sup.3, and R.sup.4 are as described in the specification. These compounds are inhibitors of protein kinase, particularly inhibitors of JNK, a mammalian protein kinase involved cell proliferation, cell death and response to extracellular stimuli; Lck and Src kinase. The invention also relates to methods for producing these inhibitors. The invention also provides pharmaceutical compositions comprising the inhibitors of the invention and methods of utilizing those compositions in the treatment and prevention of various disorders.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application claims benefit of priority to co-pending U.S. provisional applications No. 60/283,621 filed Apr. 13, 2001, No. 60/329,440 filed Oct. 14, 2001 and No. 60/292,974 filed May 23, 2001.

---------- KWIC ---------

Summary of Invention Paragraph - BSTX (2):
[0002] The present invention relates to inhibitors of protein kinase, especially c-Jun N-terminal kinases (JNK) and the Src-family of kinases, includling Lck, which are members of the mitogen-activated protein (MAP) kinase family. JNK, Src, and Lck have been implicated in a number of different human diseases. The invention also provides pharmaceutical compositions comprising the inhibitors of the invention and methods of utilizing those compositions in the treatment and prevention of various disorders in which JNK, Src, and Lck play a role.

Summary of Invention Paragraph - BSTX (17):
[0016] Based on published studies, Src kinases are considered as potential therapeutic targets for various human diseases. Mice that are deficient in Src develop osteopetrosis, or bone build-up, because of depressed bone resorption by osteoclasts. This suggests that osteoporosis resulting from abnormally high bone resorption can be treated by inhibiting Src. Soriano et al., Cell, 69, 551 (1992) and Soriano et al., Cell, 64, 693 (1991).

Summary of Invention Paragraph - BSTX (18):
[0017] Suppression of arthritic bone destruction has been achieved by the overexpression of CSK in rheumatoid synoviocytes and osteoclasts. Takayanagi et al., J. Clin. Invest., 104, 137 (1999). CSK, or C-terminal Src kinase, phosphorylates and thereby inhibits Src catalytic activity. This implies that Src inhibition may prevent joint destruction that is characteristic in patients suffering from rheumatoid arthritis. Boschelli et al., Drugs of the Future 2000, 25(7), 717, (2000).

Summary of Invention Paragraph - BSTX (20):
[0019] A number of studies have linked Src expression to cancers such as colon, breast, hepatic and pancreatic cancer, certain B-cell leukemias and lymphomas. Talamonti et al., J. Clin. Invest., 91, 53 (1993); Lutz et al., Biochem. Biophys. Res. 243, 503 (1998); Rosen et al., J. Biol. Chem., 261, 13754 (1986); Bolen et al., Proc. Natl. Acad. Sci. USA, 84, 2251 (1987); Masaki et al., Hepatology, 27, 1257 (1998); Biscardi et al., Adv. Cancer Res., 76, 61 (1999); Lynch et al., Leukemia, 7, 1416 (1993). Furthermore, antisense Src expressed in ovarian and colon tumor cells has been shown to inhibit tumor growth. Wiener et al., Clin. Cancer Res., 5, 2164 (1999); Staley et al., Cell Growth Diff., 8, 269 (1997).

Summary of Invention Paragraph - BSTX (22):
[0021] Accordingly, there is still a great need to develop potent inhibitors of JNKs and Src family kinases that are useful in treating various conditions associated with JNK and Src activation.

Summary of Invention Paragraph - BSTX (27):
[0025] The compounds and pharmaceutical compositions of the present invention are useful as inhibitors of c-Jun N-terminal kinases (JNK) and Src family kinases, including Src and Lck. Thus, they are also useful in methods for treating or preventing a variety of disorders, such as heart disease, immunodeficiency disorders, inflammatory diseases, allergic diseases, autoimmune diseases, destructive bone disorders such as osteoporosis, proliferative disorders, infectious diseases and viral diseases. The compositions are also useful in methods for preventing cell death and hyperplasia and therefore may be used to treat or prevent reperfusion/ischemia in stroke, heart attacks, and organ hypoxia. The compositions are also useful

in methods for preventing thrombin-induced platelet aggregation. The compositions are especially useful for disorders such as chronic myelogenous leukemia (CML), rheumatoid arthritis, asthma, osteoarthritis, ischemia, cancer, liver disease including hepatic ischemia, heart disease such as myocardial infarction and congestive heart failure, pathologic immune conditions involving T cell activation and neurodegenerative disorders.

Summary of Invention Paragraph - BSTX (185):
[0182] According to another embodiment, the invention provides a method of inhibiting JNK, Src, or Lck kinase activity in a biological sample. This method comprises the step of contacting said biological sample with a compound of formula I. According to a preferred embodiment, the invention relates to a method of inhibiting JNK, Src, or Lck kinase activity in a biological sample comprising the step of contacting said biological sample with a compound of formula IIa, IIb, V, or VI. A more preferred embodiment relates to contacting said biological sample with a compound of formula IIa or VI.

Summary of Invention Paragraph - BSTX (187):
[0184] Inhibition of JNK, Src, or Lck kinase activity in a biological sample is useful for a variety of purposes which are known to one of skill in the art. Examples of such purposes include, but are not limited to, blood transfusion, organ-translplantation, biological specimen storage, and biological assays.

Summary of Invention Paragraph - BSTX (188):
[0185] Compounds of formula I or salts thereof may be formulated into compositions. In a preferred embodiment, the composition is a pharmaceutically acceptable composition. In one embodiment, the composition comprises an amount of compound effective to inhibit a protein kinase, particularly JNK, Src, or Lck, in a biological sample or in a patient. In another embodiment, compounds of this invention and pharmaceutical compositions thereof, which comprise an amount of the compound effective to treat or prevent an JNK, Src, or Lck-mediated condition and a pharmaceutically acceptable carrier, adjuvant, or vehicle, may be formulated for administration to a patient.

Summary of Invention Paragraph - BSTX (189):
[0186] The amount effective to inhibit protein kinase, for example, JNK, Src, or Lck, is one that measurably inhibits the kinase activity where compared to the activity of the enzyme in the absence of an inhibitor. Any method may be used to determine inhibition.

Summary of Invention Paragraph - BSTX (209):
[0206] It should also be understood that a specific dosage and treatment regimen for any particular patient will depend upon a variety of factors, including the activity of the specific compound employed, the age, body weight, general health, sex, diet, time of administration, rate of excretion, drug combination, and the judgment of the treating physician and the severity of the particular disease being treated. The dosage of compound will also depend upon which particular compound is in the composition The compounds of this invention are inhibitors of JNK, Src, or Lck kinase as determined by enzymatic assay. Accordingly, these compounds are useful for treating JNK-, Src-, or Lck-mediated diseases or conditions.

Summary of Invention Paragraph - BSTX (212):
[0209] The activity of the compounds of this invention as kinase inhibitors may be assayed in vitro, in vivo or in a cell line. In vitro assays include assays that determine inhibition of either the kinase activity or ATPase activity of activated enzyme, for example JNK, Lck, or Src. Alternate in vitro assays quantitate the ability of the inhibitor to bind to JNK, Lck, or Src and may be measured either by radiolabelling the inhibitor prior to binding,

isolating the inhibitor/JNK, inhibitor/Lck, or inhibitor/Src complex and determining the amount of radiolabel bound, or by running a competition experiment where new compounds are incubated with JNK, Lck, or Src bound to known radioligands. One may use any type or isoform of JNK, Lck, or Src, depending upon which JNK, Lck, or Src type or isoform is to be inhibited. The details of the conditions used for the enzymatic assays are set forth in the Examples hereinbelow.

Summary of Invention Paragraph - BSTX (222):
[0219] In addition, JNK inhibitors of the present invention may be capable of inhibiting the expression of inducible pro-inflammatory proteins. Therefore, other "JNK-mediated diseases" or "conditions" which may be treated by the compounds of this invention include edema, analgesia, fever and pain, such as neuromuscular pain, headache, cancer pain, dental pain and arthritis pain.

Summary of Invention Paragraph - BSTX (223):
[0220] The compounds of this invention are also useful as inhibitors of Src-family kinases, especially Src. For a general review of these kinases see Thomas and Brugge, Annu. Rev. Cell Dev. Biol. (1997) 13, 513; Lawrence and Niu, Pharmacol. Ther. (1998) 77, 81; Tatosyan and Mizenina, Biochemistry (Moscow) (2000) 65, 49. Accordingly, these compounds are useful for treating Src-mediated diseases or conditions.

Summary of Invention Paragraph - BSTX (235):
[0232] Each of the aforementioned methods directed to the inhibition of JNK, Lck, or Src, or the treatment of a. disease alleviated thereby, is preferably carried out with a preferred compound of formula I, IIa, V, or VI, as described above. More preferably, each of the aforementioned methods is carried out with a preferred compound of formula I, IIa, V, or VI, and most preferably with a compound of formula IIa, V, or VI.

Detail Description Paragraph - DETX (138):
[0328] The following examples demonstrate how the compounds of this invention may be tested as inhibitors of c-Jun-N-terminal, Src, and Lck kinases.

Detail Description Paragraph - DETX (154):
[0337] The compounds were evaluated as inhibitors of human Src kinase using either a radioactivity-based assay or spectrophotometric assay.

Detail Description Paragraph - DETX (155):
Src Inhibition Assay A: Radioactivity-Based Assay

Detail Description Paragraph - DETX (156):
[0338] The compounds were assayed as inhibitors of full length recombinant human Src kinase (from Upstate Biotechnology, cat. no. 14-117) expressed and purified from baculo viral cells. Src kinase activity was monitored by following the incorporation of .sup.33P from ATP into the tyrosine of a random poly Glu-Tyr polymer substrate of composition, Glu:Tyr=4:1 (Sigma, cat. no. P-0275). The following were the final concentrations of the assay components: 0.05 M HEPES, pH 7.6, 10 mM MgCl.sub.2, 2 mM DTT, 0.25 mg/ml BSA, 10 pM ATP (1-2 .mu.Ci .sup.33P-ATP per reaction), 5 mg/ml poly Glu-Tyr, and 1-2 units of recombinant human Src kinase. In a typical assay, all the reaction components with the exception of ATP were pre-mixed and aliquoted into assay plate wells. Inhibitors dissolved in DMSO were added to the wells to give a final DMSO concentration of 2.5%. The assay plate was incubated at 30.degree. C. for 10 min before initiating the reaction with .sup.33P-ATP. After 20 min of reaction, the reactions were quenched with 150 .mu.l of 10% trichloroacetic

acid (TCA) containing 20 mM Na.sub.3PO.sub.4. The quenched samples were then transferred to a 96-well filter plate (Whatman, UNI-Filter GF/F Glass Fiber Filter, cat no. 7700-3310) installed on a filter plate vacuum manifold. Filter plates were washed four times with 10% TCA containing 20 mM Na.sub.3PO.sub.4 and then 4 times with methanol. 200 .mu.l of scintillation fluid was then added to each well. The plates were sealed and the amount of radioactivity associated with the filters was quantified on a TopCount scintillation counter. The radioactivity incorporated was plotted as a function of the inhibitor concentration. The data was fitted to a competitive inhibition kinetics model to get the K.sub.i for the compound.

Detail Description Paragraph - DETX (157):
Src Inhibition Assay B: Spectrophotometric Assay

Detail Description Paragraph - DETX (161):
[0342] Table 14 shows the results of the activity of selected compounds of this invention in the Src inhibition assay. The compound numbers correspond to the compound numbers in Tables 1, 2, 7, and 8. Compounds having a K.sub.i less than 0.1 micromolar (.mu.M) are rated "A", compounds having a K.sub.i between 0.1 and 1 .mu.M are rated "B" and compounds having a K.sub.i greater than 1 .mu.M are rated "C". Compounds having an activity designated as "D" provided a percent inhibition less than or equal to 24%; compounds having an activity designated as "E" provided a percent inhibition of between 24% and 66%; and compounds having an activity designated as "F" provided a provided a percent inhibition of between 67% and 100%.

Claims Text - CLTX (20):
19. A method of inhibiting JNK, Lck, or Src kinase activity in a biological sample comprising the step of contacting said biological sample with: a) a compound according to claim 1; or b) a composition according to claim 17.

PGPUB-DOCUMENT-NUMBER: 20040006083

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040006083 A1

TITLE: Pyrazolopyrimidines as therapeutic agents

PUBLICATION-DATE: January 8, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Hirst, Gavin C.	Marlborough	MA	US	
Rafferty, Paul	Westborough	MA	US	
Ritter, Kurt	Frankfurt	MA	DE	
Calderwood, David	Framingham	MA	US	
Wishart, Neil	Holden	MA	US	
Arnold, Lee D.	Westborough	MA	US	
Friedman, Michael M.	Newton		US	

APPL-NO: 10/ 104140

DATE FILED: July 19, 2002

RELATED-US-APPL-DATA:

non-provisional-of-provisional 60278047 20010322 US

US-CL-CURRENT: 514/248, 514/249 , 514/259.1 , 514/264.1 , 544/235 , 544/262

ABSTRACT:

The present invention provides compounds of Formula I, 1
including pharmaceutically acceptable salts and/or prodrugs thereof, where G, R.sub.a, R.sub.2, and R.sub.3 are defined as described herein.

---------- KWIC ---------

Summary of Invention Paragraph - BSTX (10):
[0009] As previously stated, recent evidence suggests that VEGF plays a role in the stimulation of both normal and pathological angiogenesis (Jakeman et al., Endocrinology 133: 848-859, 1993; Kolch et al., Breast Cancer Research and Treatment 36: 139-155, 1995; Ferrara et al., Endocrine Reviews 18(1); 4-25, 1997; Ferrara et al., Regulation of Angiogenesis (ed. L. D. Goldberg and E. M. Rosen), 209-232, 1997). In addition, VEGF has been implicated in the control and enhancement of vascular permeability (Connolly, et al., J. Biol. Chem. 264: 20017-20024, 1989; Brown et al., Regulation of Angiogenesis (ed. L. D. Goldberg and E. M. Rosen), 233-269, 1997). Different forms of VEGF arising from alternative splicing of mRNA have been reported, including the four species described by Ferrara et al. (J. Cell. Biochem. 47:211-218, 1991). Both secreted and predominantly cell-associated species of VEGF have been identified by Ferrara et al. supra, and the protein is known to exist in the form of disulfide linked dimers.

Summary of Invention Paragraph - BSTX (12):
[0011] Placenta growth factor (PlGF) has an amino acid sequence that exhibits significant homology to the VEGF sequence (Park et al., J. Biol.

Chem. 269:25646-54, 1994; Maglione et al. Oncogene 8:925-31, 1993). As with VEGF, different species of PlGF arise from alternative splicing of mRNA, and the protein exists in dimeric form (Park et al., supra). PlGF-1 and PlGF-2 bind to Flt-1 with high affinity, and PlGF-2 also avidly binds to neuropilin-1 (Migdal et al, J. Biol. Chem. 273 (35): 22272-22278), but neither binds to FLK-1/KDR (Park et al., supra). PlGF has been reported to potentiate both the vascular permeability and mitogenic effect of VEGF on endothelial cells when VEGF is present at low concentrations (purportedly due to heterodimer formation) (Park et al., supra).

Summary of Invention Paragraph - BSTX (16):

[0015] As for VEGF, VEGF-C and VEGF-D have been claimed to induce increases in vascular permeability in vivo in a Miles assay when injected into cutaneous tissue (PCT/US97/14696; WO98/07832, Witzenbichler et al., supra). The physiological role and significance of these ligands in modulating vascular hyperpermeability and endothelial responses in tissues where they are expressed remains uncertain.

Summary of Invention Paragraph - BSTX (18):

[0017] Based upon emerging discoveries of other homologs of VEGF and VEGFRs and the precedents for ligand and receptor heterodimerization, the actions of such VEGF homologs may involve formation of VEGF ligand heterodimers, and/or heterodimerization of receptors, or binding to a yet undiscovered VEGFR (Witzenbichler et al., supra). Also, recent reports suggest neuropilin-1 (Migdal et al, supra) or VEGFR-3/Flt-4 (Witzenbichler et al., supra), or receptors other than KDR/VEGFR-2 may be involved in the induction of vascular permeability (Stacker, S. A., Vitali, A., Domagala, T., Nice, E., and Wilks, A. F., "Angiogenesis and Cancer" Conference, Amer. Assoc. Cancer Res., January 1998, Orlando, Fla.; Williams, Diabetelogia 40: S118-120 (1997)).

Summary of Invention Paragraph - BSTX (25):

[0024] More recently, attempts have been made to identify small molecules which act as tyrosine kinase inhibitors. For example, bis monocyclic, bicyclic or heterocyclic aryl compounds (PCT WO 92/20642) and vinylene-azaindole derivatives (PCT WO 94/14808) have been described generally as tyrosine kinase inhibitors. Styryl compounds (U.S. Pat. No. 5,217,999), styryl-substituted pyridyl compounds (U.S. Pat. No. 5,302,606), certain quinazoline derivatives (EP Application No. 0 566 266 A1; Expert Opin. Ther. Pat. (1998), 8(4): 475-478), selenoindoles and selenides (PCT WO 94/03427), tricyclic polyhydroxylic compounds (PCT WO 92/21660) and benzylphosphonic acid compounds (PCT WO 91/15495) have been described as compounds for use as tyrosine kinase inhibitors for use in the treatment of cancer. Anilinocinnolines (PCT WO97/34876) and quinazoline derivative compounds (PCT WO97/22596; PCT WO97/42187) have been described as inhibitors of angiogenesis and vascular permeability.

Summary of Invention Paragraph - BSTX (26):

[0025] In addition, attempts have been made to identify small molecules which act as serine/threonine kinase inhibitors. For example, bis(indolylmaleimide) compounds have been described as inhibiting particular PKC serine/threonine kinase isoforms whose signal transducing function is associated with altered vascular permeability in VEGF-related diseases (PCT WO97/40830; PCT WO97/40831).

Summary of Invention Paragraph - BSTX (32):

[0031] Inhibitors of kinases involved in mediating or maintaining disease states represent novel therapies for these disorders. Examples of such kinases include, but are not limited to: (1) inhibition of c-Src (Brickell, Critical Reviews in Oncogenesis, 3:401-406 (1992); Courtneidge, Seminars in Cancer

Biology, 5:236-246 (1994), raf (Powis, Pharmacology & Therapeutics, 62:57-95 (1994)) and the cyclin-dependent kinases (CDKs) 1, 2 and 4 in cancer (Pines, Current Opinion in Cell Biology, 4:144-148 (1992); Lees, Current Opinion in Cell Biology, 7:773-780 (1995); Hunter and Pines, Cell, 79:573-582 (1994)), (2) inhibition of CDK2 or PDGF-R kinase in restenosis (Buchdunger et al., Proceedings of the National Academy of Science USA, 92:2258-2262 (1995)), (3) inhibition of CDK5 and GSK3 kinases in Alzheimers (Hosoi et al., Journal of Biochemistry (Tokyo), 117:741-749 (1995); Aplin et al., Journal of Neurochemistry, 67:699-707 (1996), (4) inhibition of c-Src kinase in osteoporosis (Tanaka et al., Nature, 383:528-531 (1996), (5) inhibition of GSK-3 kinase in type-2 diabetes (Borthwick et al., Biochemical & Biophysical Research Communications, 210:738-745 (1995), (6) inhibition of the p38 kinase in inflammation (Badger et al., The Journal of Pharmacology and Experimental Therapeutics, 279:1453-1461 (1996)), (7) inhibition of VEGF-R 1-3 and TIE-1 and -2 kinases in diseases which involve angiogenesis (Shawver et al., Drug Discovery Today, 2:50-63 (1997)), (8) inhibition of UL97 kinase in viral infections (He et al., Journal of Virology, 71:405-411 (1997)), (9) inhibition of CSF-1R kinase in bone and hematopoetic diseases (Myers et al., Bioorganic & Medicinal Chemistry Letters, 7:421-424 (1997), and (10) inhibition of Lck kinase in autoimmune diseases and transplant rejection (Myers et al., Bioorganic & Medicinal Chemistry Letters, 7:417-420 (1997).

Summary of Invention Paragraph - BSTX (34):

[0033] The identification of effective small compounds which specifically inhibit signal transduction and cellular proliferation by modulating the activity of receptor and non-receptor tyrosine and serine/threonine kinases to regulate and modulate abnormal or inappropriate cell proliferation, differentiation, or metabolism is therefore desirable. In particular, the identification of methods and compounds that specifically inhibit the function of a tyrosine kinase which is essential for antiogenic processes or the formation of vascular hyperpermeability leading to edema, ascites, effusions, exudates, and macromolecular extravasation and matrix deposition as well as associated disorders would be beneficial.

Summary of Invention Paragraph - BSTX (318):

[0316] In another aspect the present invention is directed to a method of treating a condition in a patient comprising administering a therapeutically effective amount of a compound of Formula (I) or a physiologically acceptable salt, prodrug or biologically active metabolites thereof to said patient, wherein said condition is an ocular condition, a cardiovascular condition, a cancer, Crow-Fukase (POEMS) syndrome, a diabetic condition, sickle cell anaemia, chronic inflammation, systemic lupus, glomerulonephritis, synovitis, inflammatory bowel disease, Crohn's disease, glomerulonephritis, rheumatoid arthritis, osteoarthritis, multiple sclerosis, graft rejection, Lyme disease, sepsis, von Hippel Lindau disease, pemphigoid, psoriasis, Paget's disease, polycystic kidney disease, fibrosis, sarcoidosis, cirrhosis, thyroiditis, hyperviscosity syndrome, Osler-Weber-Rendu disease, chronic occlusive pulmonary disease, asthma or edema following burns, trauma, radiation, stroke, hypoxia, ischemia, ovarian hyperstimulation syndrome, preeclampsia, menometrorrhagia, endometriosis, or infection by Herpes simplex, Herpes Zoster, human immunodeficiency virus, parapoxyirus, protozoa or toxoplasmosis.

Summary of Invention Paragraph - BSTX (320):

[0318] ocular or macular edema, ocular neovascular disease, scleritis, radial keratotomy, uveitis, vitritis, myopia, optic pits, chronic retinal detachment, post-laser treatment complications, conjunctivitis, Stargardt's disease, Eales disease, retinopathy or macular degeneration;

Summary of Invention Paragraph - BSTX (355):

[0352] Further, some of these compounds can be used as active agents against burns, chronic lung disease, stroke, polyps, anaphylaxis, chronic and allergic inflammation, delayed-type hypersensitivity, ovarian hyperstimulation syndrome, brain tumor-associated cerebral edema, high-altitude, trauma or hypoxia induced cerebral or pulmonary edema, ocular and macular edema, ascites, glomerulonephritis and other diseases where vascular hyperpermeability, effusions, exudates, protein extravasation, or edema is a manifestation of the disease. The compounds will also be useful in treating disorders in which protein extravasation leads to the deposition of fibrin and extracellular matrix, promoting stromal proliferation (e.g. keloid, fibrosis, cirrhosis and carpal tunnel syndrome). Increased VEGF production potentiates inflammatory processes such as monocyte recruitment and activation. The compounds of this invention will also be useful in treating inflammatory disorders such as inflammatory bowel disease (IBD) and Crohn's disease.

Summary of Invention Paragraph - BSTX (368):

[0365] VEGF's are unique in that they are the only angiogenic growth factors known to contribute to vascular hyperpermeability and the formation of edema. Indeed, vascular hyperpermeability and edema that is associated with the expression or administration of many other growth factors appears to be mediated via VEGF production. Inflammatory cytokines stimulate VEGF production. Hypoxia results in a marked upregulation of VEGF in numerous tissues, hence situations involving infarct, occlusion, ischemnia, anemia, or circulatory impairment typically invoke VEGF/VPF mediated responses. Vascular hyperpermeability, associated edema, altered transendothelial exchange and macromolecular extravasation, which is often accompanied by diapedesis, can result in excessive matrix deposition, aberrant stromal proliferation, fibrosis, etc. Hence, VEGF-mediated hyperpermeability can significantly contribute to disorders with these etiologic features.

Summary of Invention Paragraph - BSTX (371):

[0368] The compounds of this invention have inhibitory activity against protein kinases. That is, these compounds modulate signal transduction by protein kinases. Compounds of this invention inhibit protein kinases from serine/threonine and tyrosine kinase classes. In particular, these compounds selectively inhibit the activity of the KDR/FLK-1/VEGFR-2 tyrosine kinases. Certain compounds of this invention also inhibit the activity of additional tyrosine kinases such as Flt-1/VEGFR-1, Flt-4, Tie-1, Tie-2, FGFR, PDGFR, IGF-1R, c-Met, Src-subfamily kinases such as Lck, Src, hck, fgr, fyn, yes, etc. Additionally, some compounds of this invention significantly inhibit serine/threonine kinases such as PKC, MAP kinases, erk, CDKs, Plk-1, or Raf-1 which play an essential role in cell proliferation and cell-cycle progression. The potency and specificity of the generic compounds of this invention towards a particular protein kinase can often be altered and optimized by variations in the nature, number and arrangement of the substituents (i.e., R.sub.1, R.sub.2, R.sub.3, A and ring 1) and conformational restrictions. In addition the metabolites of certain compounds may also possess significant protein kinase inhibitory activity.

Summary of Invention Paragraph - BSTX (372):

[0369] The compounds of this invention, when administered to individuals in need of such compounds, inhibit vascular hyperpermeability and the formation of edema in these individuals. These compounds act, it is believed, by inhibiting the activity of KDR tyrosine kinase which is involved in the process of vascular hyperpermeability and edema formation. The KDR tyrosine kinase may also be referred to as FLK-1 tyrosine kinase, NYK tyrosine kinase or VEGFR-2 tyrosine kinase. KDR tyrosine kinase is activated when vascular endothelial cell growth factor (VEGF) or another activating ligand (such as VEGF-C, VEGF-D, VEGF-E or HIV Tat protein) binds to a KDR tyrosine kinase receptor which lies

on the surface of vascular endothelial cells. Following such KDR tyrosine kinase activation, hyperpermeability of the blood vessels occurs and fluid moves from the blood stream past the blood vessel walls into the interstitial spaces, thereby forming an area of edema. Diapedesis also often accompanies this response. Similarly, excessive vascular hyperpermeability can disrupt normal molecular exchange across the endothelium in critical tissues and organs (e.g., lung and kidney), thereby causing macromolecular extravasation and deposition. Following this acute response to KDR stimulation which is believed to facilitate the subsequent angiogenic process, prolonged KDR tyrosine kinase stimulation results in the proliferation and chemotaxis of vascular endothelial cells and formation of new vessels. By inhibiting KDR tyrosine kinase activity, either by blocking the production of the activating ligand, by blocking the activating ligand binding to the KDR tyrosine kinase receptor, by preventing receptor dimerization and transphosphorylation, by inhibiting the enzyme activity of the KDR tyrosine kinase (inhibiting the phosphorylation function of the enzyme) or by some other mechanism that interrupts its downstream signaling (D. Mukhopedhyay et al., Cancer Res. 58:1278-1284 (1998) and references therein), hyperpermeability, as well as associated extravasation, subsequent edema formation and matrix deposition, and angiogenic responses, may be inhibited and minimized.

Summary of Invention Paragraph - BSTX (378):

[0375] The method of the present invention is useful in the treatment of protein kinase-mediated conditions, such as any of the conditions described above. In one embodiment, the protein kinase-mediated condition is characterized by undesired angiogenesis, edema, or stromal deposition. For example, the condition can be one or more more ulcers, such as ulcers caused by bacterial or fungal infections, Mooren ulcers and ulcerative colitis. The condition can also be due to a microbial infection, such as Lyme disease, sepsis, septic shock or infections by Herpes simplex, Herpes Zoster, human immunodeficincy virus, protozoa, toxoplasmosis or parapoxyirus; an angiogenic disorders, such as von Hippel Lindau disease, polycystic kidney disease, pemphigoid, Paget's disease and psoriasis; a reproductive condition, such as endometriosis, ovarian hyperstimulation syndrome, preeclampsia or menometrorrhagia; a fibrotic and edemic condition, such as sarcoidosis, fibrosis, cirrhosis, thyroiditis, hyperviscosity syndrome systemic, Osler-Weber-Rendu disease, chronic occlusive pulmonary disease, asthma, and edema following burns, trauma, radiation, stroke, hypoxia or ischemia; or an inflammatory/immunologic condition, such as systemic lupus, chronic inflammation, glomerulonephritis, synovitis, inflammatory bowel disease, Crohn's disease, rheumatoid arthritis, osteoarthritis, multiple sclerosis and graft rejection. Suitable protein kinase-mediated conditions also include sickle cell anaemia, osteoporosis, osteopetrosis, tumor-induced hypercalcemia and bone metastases. Additional protein kinase-mediated conditions which can be treated by the method of the present invention include ocular conditions such as ocular and macular edema, ocular neovascular disease, scleritis, radial keratotomy, uveitis, vitritis, myopia, optic pits, chronic retinal detachment, post-laser complications, conjunctivitis, Stargardt's disease and Eales disease, in addition to retinopathy and macular degeneration.

Summary of Invention Paragraph - BSTX (384):

[0381] In many pathological conditions (for example, solid primary tumors and metastases, Kaposi's sarcoma, rheumatoid arthritis, blindness due to inappropriate ocular neovascularization, psoriasis and atherosclerosis) disease progression is contingent upon persistent angiogenesis. Polypeptide growth factors often produced by the disease tissue or associated inflammatory cells, and their corresponding endothelial cell specific receptor tyrosine kinases (e.g., KDR/VEGFR-2, Flt-1/VEGFR-1, Flt-4, Tie-2/Tek and Tie) are essential for the stimulation of endothelial cell growth, migration, organization,

differentiation and the establishment of the requisite new functional vasculature. As a result of the vascular permeability factor activity of VEGF in mediating vascular hyperpermeability, VEGF-stimulation of a VEGFR kinase is also believed to play an important role in the formation of tumor ascites, cerebral and pulmonary edema, pleural and pericardial effusions, delayed-type hypersensitivity reactions, tissue edema and organ dysfunction following trauma, burns, ischemia, diabetic complications, endometriosis, adult respiratory distress syndrome (ARDS), post-cardiopulmonary bypass-related hypotension and hyperpermeability, and ocular edema leading to glaucoma or blindness due to inappropriate neovascularization. In addition to VEGF, recently identified VEGF-C and VEGF-D, and virally-encoded VEGF-E or HIV-Tat protein can also cause a vascular hyperpermeability response through the stimulation of a VEGFR kinase. KDR/VEGFR-2 and/or Tie-2 are expressed also in a select population of hematopoietic stem cells. Certain members of this population are pluripotent in nature and can be stimulated with growth factors to differentiate into endothelial cells and participate in vasculogenetic angiogenic processes. For this reason these have been called Endothelial Progenitor Cells (EPCs) (J. Clin. Investig. 103: 1231-1236 (1999)). In some progenitors, Tie-2 may play a role in their recruitment, adhesion, regulation and differentiation (Blood, 4317-4326 (1997)). Certain agents according to formula I capable of blocking the kinase activity of endothelial cell specific kinases could therefore inhibit disease progression involving these situations.

Summary of Invention Paragraph - BSTX (386):

[0383] The compounds of formula I or a salt thereof or pharmaceutical compositions containing a therapeutically effective amount thereof may be used in the treatment of protein kinase-mediated conditions, such as benign and neoplastic proliferative diseases and disorders of the immune system, as described above. For example, such diseases include autoimmune diseases, such as rheumatoid arthritis, thyroiditis, type 1 diabetes, multiple sclerosis, sarcoidosis, inflammatory bowel disease, Crohn's disease, myasthenia gravis and systemic lupus erythematosus; psoriasis, organ transplant rejection (eg. kidney rejection, graft versus host disease), benign and neoplastic proliferative diseases, human cancers such as lung, breast, stomach, bladder, colon, pancreas, ovarian, prostate and rectal cancer and hematopoietic malignancies (leukemia and lymphoma), and diseases involving inappropriate vascularization for example diabetic retinopathy, retinopathy of prematurity, choroidal neovascularization due to age-related macular degeneration, and infantile hemangiomas in human beings. In addition, such inhibitors may be useful in the treatment of disorders involving VEGF mediated edema, ascites, effusions, and exudates, including for example macular edema, cerebral edema, acute lung injury and adult respiratory distress syndrome (ARDS).

Summary of Invention Paragraph - BSTX (393):

[0390] The compounds of this invention can be administered to a human patient by themselves or in pharmaceutical compositions where they are mixed with suitable carriers or excipient(s) at doses to treat or ameliorate vascular hyperpermeability, edema and associated disorders. Mixtures of these compounds can also be administered to the patient as a simple mixture or in suitable formulated pharmaceutical compositions. A therapeutically effective dose further refers to that amount of the compound or compounds sufficient to result in the prevention or attenuation of inappropriate neovascularization, progression of hyperproliferative disorders, edema, VEGF-associated hyperpermeability and/or VEGF-related hypotension. Techniques for formulation and administration of the compounds of the instant application may be found in "Remington's Pharmaceutical Sciences," Mack Publishing Co., Easton, Pa., latest edition.

Summary of Invention Paragraph - BSTX (396):

[0393] Alternatively, one may administer the compound in a local rather than a systemic manner, for example, via injection of the compound directly into an edematous site, often in a depot or sustained release formulation.

Summary of Invention Paragraph - BSTX (434):

[0431] In the compositions of the present invention the active compound may, if desired, be associated with other compatible pharmacologically active ingredients. For example, the compounds of this invention can be administered in combination with one or more additional pharmaceutical agents that inhibit or prevent the production of VEGF or angiopoietins, attenuate intracellular responses to VEGF or angiopoietins, block intracellular signal transduction, inhibit vascular hyperpermeability, reduce inflammation, or inhibit or prevent the formation of edema or neovascularization. The compounds of the invention can be administered prior to, subsequent to or simultaneously with the additional pharmaceutical agent, whichever course of administration is appropriate. The additional pharmaceutical agents include but are not limited to anti-edemic steroids, NSAIDS, ras inhibitors, anti-TNF agents, anti-ILI agents, antihistamines, PAF-antagonists, COX-1 inhibitors, COX-2 inhibitors, NO synthase inhibitors, Akt/PTB inhibitors, IGF-1R inhibitors, PKC inhibitors and P13 kinase inhibitors. The compounds of the invention and the additional pharmaceutical agents act either additively or synergistically. Thus, the administration of such a combination of substances that inhibit angiogenesis, vascular hyperpermeability and/or inhibit the formation of edema can provide greater relief from the deletrious effects of a hyperproliferative disorder, angiogenesis, vascular hyperpermeability or edema than the administration of either substance alone. In the treatment of malignant disorders combinations with antiproliferative or cytotoxic chemotherapies or radiation are anticipated.

Summary of Invention Paragraph - BSTX (525):

[0522] In vivo Uterine Edema Model

Summary of Invention Paragraph - BSTX (526):

[0523] This assay measures the capacity of compounds to inhibit the acute increase in uterine weight in mice which occurs in the first few hours following estrogen stimulation. This early onset of uterine weight increase is known to be due to edema caused by increased permeability of uterine vasculature. Cullinan-Bove and Koss (Endocrinology (1993), 133:829-837) demonstrated a close temporal relationship of estrogen-stimulated uterine edema with increased expression of VEGF mRNA in the uterus. These results have been confirmed by the use of neutralizing monoclonal antibody to VEGF which significantly reduced the acute increase in uterine weight following estrogen stimulation (WO 97/42187). Hence, this system can serve as a model for in vivo inhibition of VEGF signalling and the associated hyperpermeability and edema.

Summary of Invention Paragraph - BSTX (535):

[0532] Results demonstrate that certain compounds of the present invention inhibit the formation of edema when administered systemically by various routes.

Claims Text - CLTX (33):

32. A method of treating a condition in a patient comprising administering a therapeutically effective amount of a compound of claim 1 or a physiologically acceptable salt, prodrug or biologically active metabolites thereof to said patient, wherein said condition is an ocular condition, a cardiovascular condition, a cancer, Crow-Fukase (POEMS) syndrome, a diabetic condition, sickle cell anaemia, chronic inflammation, systemic lupus, glomerulonephritis, synovitis, inflammatory bowel disease, Crohn's disease, glomerulonephritis, rheumatoid arthritis, osteoarthritis, multiple sclerosis,

graft rejection, Lyme disease, sepsis, von Hippel Lindau disease, pemphigoid, psoriasis, Paget's disease, polycystic kidney disease, fibrosis, sarcoidosis, cirrhosis, thyroiditis, hyperviscosity syndrome, Osler-Weber-Rendu disease, chronic occlusive pulmonary disease, asthma or edema following burns, trauma, radiation, stroke, hypoxia, ischemia, ovarian hyperstimulation syndrome, preeclampsia, menometrorrhagia, endometriosis, or infection by Herpes simplex, Herpes Zoster, human immunodeficiency virus, parapoxyirus, protozoa or toxoplasmosis.

Claims Text - CLTX (34):

33. The method of claim 32 wherein the ocular condition is ocular or macular edema, ocular neovascular disease, scleritis, radial keratotomy, uveitis, vitritis, myopia, optic pits, chronic retinal detachment, post-laser treatment complications, conjunctivitis, Stargardt's disease, Eales disease, retinopathy or macular degeneration.

PGPUB-DOCUMENT-NUMBER: 20040002479

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040002479 A1

TITLE: Peptide analogues and uses thereof

PUBLICATION-DATE: January 1, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Wang, Yihan	Newton	MA	US	
Sawyer, Tomi K.	Southborough	MA	US	
Berstein, David L.	Waban	MA	US	

APPL-NO: 10/ 386649

DATE FILED: March 12, 2003

RELATED-US-APPL-DATA:

non-provisional-of-provisional 60363961 20020312 US

US-CL-CURRENT: 514/79, 514/114 , 514/89 , 546/22 , 548/413 , 558/168

ABSTRACT:

This invention relates to compounds of the general formula: 1
in which R.sup.A, R.sup.B, R.sup.C, R.sup.D, R.sup.G and Z are as defined herein, and to their preparation and use.

---------- KWIC ---------

Summary of Invention Paragraph - BSTX (185):
[0184] It will be appreciated that the compounds, as described herein, may be substituted with any number of substituents or functional moieties. In general, the term "substituted" whether preceded by the term "optionally" or not, and substituents contained in formulas of this invention, refer to the replacement of hydrogen radicals in a given structure with the radical of a specified substituent. When more than one position in any given structure may be substituted with more than one substituent selected from a specified group, the substituent may be either the same or different at every position. As used herein, the term "substituted" is contemplated to include all permissible substituents of organic compounds. In a broad aspect, the permissible substituents include acyclic and cyclic, branched and unbranched, carbocyclic and heterocyclic, aromatic and nonaromatic substituents of organic compounds. For purposes of this invention, heteroatoms such as nitrogen may have hydrogen substituents and/or any permissible substituents of organic compounds described herein which satisfy the valencies of the heteroatoms. Furthermore, this invention is not intended to be limited in any manner by the permissible substituents of organic compounds. Combinations of substituents and variables envisioned by this invention are preferably those that result in the formation of stable compounds useful in the treatment, for example of bone related disorders, cancer, disorders related to increases in **vascular permeability**, and/or disorders related to call signalling. The term "stable", as used herein, preferably refers to compounds which possess stability sufficient to

allow manufacture and which maintain the integrity of the compound for a sufficient period of time to be detected and preferably for a sufficient period of time to be useful for the purposes detailed herein.

Detail Description Paragraph - DETX (39):

[0280] A murine hypercalcemia model for determining the efficacy of Src kinase inhibitors was developed. This model exploits the intrinsic effects of PTH (1-34) to stimulate the resorptive activity of osteoclasts in vivo. Briefly, compounds are each injected into mice subcutaneously, once or twice per day for five consecutive days. On the third day of test compound treatments, PTH administration begins. PTH (20 .mu.g/kg) is given four times per day, subcutaneously, until the end of the study. Control animals receive PTH but do not receive test compounds. Blood samples are collected from the animals to obtain baseline (pre-PTH treatment), 48 hour and 72 hour (after initiation of PTH treatment) serum samples. The serum samples are analyzed for calcium concentration using the quantitative colorimetric assay reagent Arsenazo III (Sigma). Calcium serum levels for treated groups are compared to calcium serum levels of control groups and a percentage of inhibition of hypercalcemia is calculated for each time point. When a compound is effective in inhibiting the activity of osteoclasts, observed serum calcium concentrations are lower than those in animals that receive only PTH in the absence of test compound.

PGPUB-DOCUMENT-NUMBER: 20030229906

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20030229906 A1

TITLE: Methods and compositions for the treatment of disorders of HIV infection

PUBLICATION-DATE: December 11, 2003

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Gelman, Irwin H.	Buffalo	NY	US	
Klotman, Paul	New York	NY	US	
Zhou, Ming Ming	Old Greenwich	CT	US	

APPL-NO: 10/ 413785

DATE FILED: April 14, 2003

RELATED-US-APPL-DATA:

non-provisional-of-provisional 60372557 20020415 US

US-CL-CURRENT: 800/8, 424/145.1 , 435/325 , 435/456 , 435/5

ABSTRACT:

The present invention relates to methods and compositions for use in the intervention of diseases associated with HIV infection. In exemplary embodiments, methods and compositions for the treatment of HIV associated nephropathy (HIVAN) are disclosed.

[0001] The present application claims the benefit of priority of U.S. Provisional Application No. 60/372,557, which was filed on Apr. 15, 2002 and is incorporated herein by reference in its entirety.

---------- KWIC ---------

Summary of Invention Paragraph - BSTX (2):

[0002] The present invention generally relates to the treatment or inhibition of diseases associated with HIV-1 infection. In particular, the present invention provides methods and compositions for decreasing, inhibiting, or otherwise abrogating the interaction of Nef with a SH3 domain of a Src family tyrosine kinase.

Summary of Invention Paragraph - BSTX (6):

[0005] HIVAN is characterized by proteinuria, rapidly developing azotemia and histologically by collapsing variant of focal and segmental glomerulosclerosis with acute tubular necrosis (Rajvanshi et al., J Assoc Physicians India, 49:813-8, 2001) and proliferation of renal tubular, parietal, and visceral epithelial cells (podocytes). In addition, the disease manifests in tubulointerstitial infiltration with mononuclear cells, edema, fibrosis, and microcystic tubule dilation (D'Agati et al., Kidney Int. 35:1358-1370, 1989;

Cohen et al., Mod Pathol. 1:87-97, 1988). Untreated, it may result in end stage renal disease (ESRD) in as little as four months and is the third leading cause of ESRD in blacks age 20 to 64 (Monahan et al., Semin Nephrol., 21(4):394-402, 2001). The incidence of HIVAN continues to increase and is the single most common cause of chronic renal disease in HIV-1 seropositive patients. Improvements in survival rates of HIV-1-seropositive patients on hemodialysis and improved treatment of HIV with highly active antiretroviral therapy (HAART) and angiotensin-converting enzyme (ACE)-inhibitors will result in an increased prevalence of HIVAN in ESRD and pre-ESRD patient populations. Thus, left unchecked HIVAN promises to become an urban epidemic as anti-HIV treatments prolong the lives of HIV-infected patients.

Summary of Invention Paragraph - BSTX (11):
[0009] The present invention provides methods of inhibiting kidney cell dedifferentiation, comprising inhibiting the interaction of Nef with a Src family tyrosine kinase SH3 domain of a polypeptide of the cell. Such a cell may be located in vitro or in vivo. In preferred embodiments, the Nef is HIV-1 Nef. In particularly preferred embodiments, the kidney cell is a podocyte.

Summary of Invention Paragraph - BSTX (12):
[0010] In general terms, inhibiting the interaction of Nef with a SH3 domain of a Src family tyrosine kinase comprises reducing the expression of Nef in the cell. In certain embodiments in which it is desirable to reduce the expression of Nef, the method comprises contacting the cell with a nucleic acid construct that reduces the expression of Nef in the cell.

Summary of Invention Paragraph - BSTX (13):
[0011] In other aspects, inhibiting the interaction of Nef with a SH3 domain of a Src family tyrosine kinase comprises contacting the Nef with an agent that binds to and/or inactivates the Nef. In specific embodiments, the agent may be a peptide inhibitor comprising a variant of the PXXP motif of the SH3 binding domain of Nef. Exemplary peptide inhibitors may comprise a sequence selected from the group consisting of SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ 4, SEQ ID NO:5, SEQ ID NO: 6, SEQ ID NO:7, SEQ ID NO:8, SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:13, SEQ ID NO:14, and SEQ ID NO:15. Of course it should be understood that these inhibitors are merely exemplary and given the teachings of the present invention, those of skill in the art will be able to design other peptide inhibitors that binds to and/or inactivates the Nef.

Summary of Invention Paragraph - BSTX (27):
[0025] Also described is a method of treating a subject, comprising inhibiting the interaction of Nef with a SH3 domain of a Src family tyrosine kinase, wherein the subject has a disease associated with HIV-1 infection. Exemplary diseases associated HIV-1 infection include but are not limited to a HIV-induced disease selected from the group consisting of HIV associated nephropathy (HIVAN) AIDS dementia; anemia; lymphoma; myopathy; cardiomyopathy; and primary HIV-induced disease progression. Contemplated treatment methods include administering compositions identified according to the present invention, either alone and/or in combination with other anti-HIV treatments.

Detail Description Paragraph - DETX (5):
[0045] In light of the above findings, the present invention provides methods and compositions for the treatment disorders associated with HIV infection. In particular, the methods and compositions are designed to disrupt, inhibit, decrease or otherwise abrogate the interaction of Nef with the SH3 domain of Src family tyrosine kinases. Such methods and compositions antagonize Nef's ability to activate proliferation and dedifferentiation signaling mediated by the Src family tyrosine kinases. The invention includes

methods of making and using peptides, small molecule inhibitors, nucleic acid-based therapies either alone or in combination with other known therapeutic interventions for HIV-based disorders. It should be noted that while the Examples of the present invention are written with respect to HIVAN, the methods and compositions of the present invention may be used in the therapeutic intervention of any disorder associated with HIV infection that is mediated through a Nef/Src interaction. The methods and compositions of the present invention are described in further detail herein below.

Detail Description Paragraph - DETX (10):

[0050] Treatment of HIVAN and such other secondary diseases that result from the expression of HIV-1 Nef in non-lymphoid tissues can be achieved through the inhibition, ablation, depletion or other reduction of the Nef/Src interaction. As such, the present invention contemplates the use of small molecules, peptides, peptidomimetic, antibodies, nucleic acids to disrupt the interaction of Nef/Src and thereby effect a beneficial outcome. Methods and compositions for achieving such a beneficial outcome are described in greater detail herein below.

Detail Description Paragraph - DETX (17):

[0057] The inventors have shown that the expression of HIV-1 Nef in non-lymphoid cells infected with HIV-1 results in the expression of genes that are deleterious to the normal phenotype. In an exemplary embodiment, it is shown herein that the nephropathy seen in HIV infection is caused by an expression of HIV-1 Nef in the podocytes. The Nef, through an interaction with the SH3 binding domain of Src family tyrosine kinases, induces dedifferentiation of the podocytes. This dedifferentiation mimics the dedifferentiation of podocytes seen in HIV-infected subjects. Taking these findings into account, the present invention is directed to methods and compositions for the treatment of disorders that appear in HIV infection. These methods and compositions are designed to disrupt or interfere with the interaction of Nef with Src family tyrosine kinases. The compositions may be any composition that interferes with, and reduces, inhibits or decreases the Nef/Src tyrosine kinase interaction. As such, the present invention specifically contemplates peptides, small molecule inhibitors, anti-Nef antibodies, peptidomimetics, antisense nef nucleic acids, and the like.

Detail Description Paragraph - DETX (23):

[0063] The present invention provides peptides that may be used as inhibitors of the interaction of Nef with Src family tyrosine kinases. Exemplary peptides include:

Detail Description Paragraph - DETX (28):

[0068] In the above table, the amino acids of the primary sequence is comprised of 16 amino acids. In the following description, the residues at each of the positions is referred to using a "P" followed by a number. For example, the primary sequence is P.sup.1P.sup.2P.sup.3P.sup.4P.sup.5P-.sup.6P.sup.7P.sup.8P.sup.9P.sup.10P.sup.12P.sup.3P.sup.14P.sup.15P.sup.16 in which P.sup.1 is G, P.sup.3 is F, P.sup.4 is P, P.sup.5 is V, P.sup.6 is R, P.sup.7 is P, P.sup.8 is Q, P.sup.9 is V, P.sup.10 is P, P.sup.11 is L, P.sup.12 is R, P.sup.13 is P, P.sup.14 is M, P.sup.15 is T, and P.sup.16 is Y. The peptides may be designed to comprise a sequence that increases the uptake of the peptides. For example, the peptides may comprise an N-terminal sequence that enhances the permeability of the molecule. In the peptides of the present invention that are contemplated to be useful as inhibitors of the Nef/Src interaction, peptides which comprise V or I in P.sup.1 are particularly preferred; at P.sup.2, the amino acid residue is preferably G; the amino acid residue at P.sup.3 may preferably be F or it also may be V; P.sup.4 may comprise P, S, Y, R or A; at P.sup.5 the residue is preferably V but also may

be A; P.sup.6 is preferably R but also may be T, K, A, M, W, S, H, Q, or C; P.sup.7 required to be P; P.sup.8 is preferably Q also may be K, R, N, G, C, or Q; P.sup.9 in preferred peptides is V, but also may be T, L or any non-polar residue; P.sup.10 is required to be P required; P.sup.11 in preferred peptides is L; in preferred P.sup.12 is R; P.sup.13 is preferably P but also may be A, E, T, S, P, I or Q; M in P.sup.14 is preferred but P.sup.14 also may be I or L; T in P15 is preferred but P.sup.15 also may be S or A; Y in P.sup.16 is preferred but P.sup.16 also may be R, H or F. One of skill in the art will be able to construct numerous variations of peptides for use in the present invention using these preferred amino acids at the indicated positions.

Detail Description Paragraph - DETX (31):

[0071] The peptide inhibitors of the present invention may be any length of amino acids so long as the amino acids are of a sufficient length to interfere with the interaction of Nef with a Src family tyrosine kinase (referred to herein as Nef/Src interaction). Preferably, the novel peptide inhibitors of the Nef/Src interaction are at least about five amino acids in length, in certain embodiments the novel peptides of the present invention may comprise a contiguous amino acid sequence of about 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 45, 50, or more amino acids.

Detail Description Paragraph - DETX (32):

[0072] In considering the particular amino acid to be positioned at any of the positions of the peptide inhibitors, it may be useful to consider the hydropathic index of amino acids at each of the positions in a peptide known to be-an effective inhibitor of the Nef binding to the SH3 domain of a Src family tyrosine kinase, and substitute a given amino acid with one of a similar hydropathic index. It is accepted that the relative hydropathic character of the amino acid contributes to the secondary structure of a resultant protein or peptide, which in turn defines the interaction of that protein with other molecules, for example, enzymes, substrates, receptors, DNA, antibodies, antigens, and the like. Each amino acid has been assigned a hydropathic index on the basis of their hydrophobicity and charge characteristics (Kyte & Doolittle, J. Mol. Biol., 157(1):105-132, 1982, incorporated herein by reference). Generally, amino acids may be substituted by other amino acids that have a similar hydropathic index or score and still result in a protein with similar biological activity i.e., still obtain a biological functionally equivalent protein or peptide. In the context of the peptides of the present invention, a biologically functionally equivalent protein or peptide will be one which still retains its ability to be an antagonist of the Nef binding to a SH3 domain of a Src family tyrosine kinase.

Detail Description Paragraph - DETX (53):

[0093] With respect to small molecule inhibitors such compounds may be identified through standard screening assays. For example, it is known that Nef interacts with Src family tyrosine kinases through binding to the SH3 domain of such kinases. Various candidate substances can be contacted with Nef followed by further determination of the ability of treated Nef to bind to an SH3 domain of a Src family tyrosine kinase. An agent that inhibits such binding will be a useful for blocking the Nef/Src interaction. Small molecule inhibitors of Nef expression that inhibit LTR-mediated transcription or splicing or Nef-specific mRNAs are contemplated to be useful in the present invention. For the former example, flavopiridol ameliorates HIVAN in transgenic mice by inhibiting cellular Cdk9, an enzyme required for HIV-1 Tat to induce LTR-mediated virus transcription. Flavopiridol, derivatives thereof and compounds in the same class as flavopiridol may be used in the present invention.

Detail Description Paragraph - DETX (57):

[0097] With respect to inhibitors that mimic Nef targets, the use of mimetics provides one example of custom designed molecules. Such molecules may be small molecule inhibitors that specifically inhibit Nef protein activity or binding to a Src family tyrosine kinase. Such molecules may be sterically similai to the actual target compounds, at least in key portions of the target's structure and or organochemical in structure. Alternatively these inhibitors may be peptidyl compounds, these are called peptidomimetics. Peptide mimetics are peptide-containing molecules which mimic elements of protein secondary structure. The underlying rationale behind the use of peptide mimetics is that the peptide backbone of proteins exists chiefly to orient amino acid side chains in such a way as to facilitate molecular interactions, such as those of ligand and receptor. An exemplary peptide mimetic of the present invention would, when administered to a subject, bind Nef in a manner analogous to the Src family tyrosine kinase SH3 domain binding to wild-type Nef.

Detail Description Paragraph - DETX (65):

[0105] The unique features of the RT-loop binding between the two proteins make this interaction an ideal target site for designing small molecular chemical inhibitors that bind selectively to this cavity in order to disrupt the interaction of Nef with the Src SH3 domain. It would be desirable to select this specific target because: (i) targeting the viral Nef protein rather than the human SH3 domain protein to block the Nef/SH3 association is preferable for therapeutic agent development, because targeting the viral protein can greatly minimize potential side effects or toxicity of drug molecules that result from their possible interferences on biochemical or biological functions of mammalian protein (such as, for example the Src-like tyrosine kinase proteins); (ii) the surface-exposed RT loop-binding site on Nef makes it readily accessible for interactions with designed chemical ligands; and (iii) the protein-protein interactions are largely hydrophobic in nature, which makes it easier to identify small molecular chemicals that bind specifically to the site. These initial binding chemical compounds can help chemical lead optimization to improve ligand binding-affinity and selectivity.

Detail Description Paragraph - DETX (68):

[0108] For a detailed description of methods for identification of small molecule inhibitors those of skill in the art are referred to WO01/51521, which describes the three-dimensional structure of a complex between phosphotyrosine binding domain of Suc 1-associated neurotrophic factor target protein and the SNT binding site of fibroblast growth factor receptor. Rational drug design predicated on the three-dimensional structure of this interaction is described in detail. It is contemplated that the techniques therein may be used for rational drug design to identify agents that can inhibit the deleterious effects of Nef binding to Src-like tyrosine kinases. For example such a method would involve identifying a compound that destabilizes the Nef/Src interaction and would involve obtaining a set of atomic coordinates that define the three dimensional structure of a Nef/Src interaction. These coordinates are determined using a complex which comprises an HIV Nef protein interacting with a Src-like tyrosine kinase (Nef/Src complex). The next step involves performing rational drug design with the atomic coordinates to select a drug that interferes with the Nef/Src complex at a given site (e.g., at the RT loop). This rational drug design is preferably performed in conjunction with computer modeling. Upon selection of the candidate drug, the candidate is contacted with a Nef/Src complex comprising a full length or fragment of Nef protein and a full length or fragment of a Src-like tyrosine kinase protein. The stability of the Nef/Src complex is monitored in the presence and absence of the candidate substance to identify a potential therapeutic agent which destabilizes the complex. Similar methods may be performed to identify a

compound which inhibits the formation of the complex. Such methods are described in detail in WO01/51521.

Detail Description Paragraph - DETX (107):

[0147] Yet another use of the Nef transgenic mouse described herein provides a new disease model for HIVAN. As shown in the data in the examples, the transgenic cells of the present invention demonstrates all the molecular and morphological features of nephropathy. A transgenic animal already exists that is being used as a model for HIVAN, but that animal expresses the entire HIV-1 genome with the exception of the gag/pol genes (Dickie et al., Virology 185:109-119, 1991; Kopp et al., Contrib Nephrol 107:194-204, 1994). Moreover, the expression of HIV-1 genes in that animal is ubiquitous. An exemplary animal of the present invention expresses far fewer genes than the gag/pol deleted mouse described by Dickie et al., (Virology 185:109-119, 1991) and Kopp et al., (Contrib Nephrol 107:194-204, 1994). In addition, another exemplary animal of the present invention expresses Nef only in kidney cells and more particularly, the animal expresses HIV-1 Nef only in the podocytes. In exemplary embodiments, expression of the Nef gene in kidney cell alone is achieved through the use of a kidney cell-specific promoter. In those embodiments where the Nef is being expressed in podocytes alone, such expression is effected through the use of a podocyte specific promoter. The nephrin promoter is one such glomerular specific promoter that may be used to target the expression of the HIV-1 Nef in the podocytes (Wong et al., Am. J. Renal. Physiol. 279:F1027-F1032, 2000). Another promoter that may be used in the present invention is beta-actin/beta-globin promoter (CX promoter) which could allow a podocyte-specific expression of a molecule of interest in kidney (Imai et al., Exp Nephrol., 7(1):63-6, 1999). In addition, the synaptopodin and podocin promoters also may be used to achieve kidney cell-specific expression. Thus, the Nef transgenic mouse provides a novel model for the study of nephropathy. This model could be exploited by treating the animal with compounds that potentially inhibit the Nef/Src interaction and treat HIV-related nephropathy. Also it is contemplated that such inhibitors may be useful in the treatment of other kidney disease as well as other disorders associated with HIV-1 infection.

Detail Description Paragraph - DETX (109):

[0149] The present invention also contemplates screening of compounds for their ability to inhibit the Nef based activation of Src family tyrosine kinases. The present invention shows that this interaction is responsible for the secondary sequelae seen in HIV infection. This realization affords the ability to create cellular, organ and organismal systems which mimic these diseases, which provide an ideal setting in which to test various compounds for therapeutic activity. Particularly preferred compounds will be those useful in inhibiting HIVAN and preventing or reversing kidney disease associated with HIV infection mediated. In the screening assays of the present invention, the candidate substance may first be screened for basic biochemical activity--e.g., binding to a target molecule--and then tested for its ability to inhibit a nephropathic phenotype, at the cellular, tissue or whole.animal level.

Detail Description Paragraph - DETX (113):

[0153] In these embodiment, the present invention is directed to a method for determining the ability of a candidate substance to inhibit the Nef/Src interaction, generally including the steps of:

Detail Description Paragraph - DETX (122):

[0162] As used herein the term "candidate substance" refers to any molecule that may potentially act as an inhibitor of the present invention. The candidate substance may be a protein or fragment thereof, a small molecule inhibitor, or even a nucleic acid molecule. It may prove to be the case that

the most useful pharmacological compounds will be compounds that are structurally related to other known modulators of HIV related nephropathy or other kidney disease, such as broad spectrum nucleoside reverse transcriptase inhibitors (e.g., Zidovudine) and other drugs commonly used in HAART, steroids such as prednisone and other corticosteroids, ramipril and the like (see Winston el al., Semin. Nephrol., 20(3):293-298, 2000 for review of treatments for HIVAN). Rational drug design includes not only comparisons with known inhibitors, but predictions relating to the structure of target molecules. Particularly useful compounds for use in rational drug design are those that will inhibit the interaction of Nef with the RT loop of the SH3 domain of Src family tyrosine kinases.

Detail Description Paragraph - DETX (126):

[0166] Candidate compounds may include fragments or parts of naturally-occurring compounds or may be found as active combinations of known compounds which are otherwise inactive. It is proposed that compounds isolated from natural sources, such as animals, bacteria, fungi, plant sources, including leaves and bark, and marine samples may be assayed as candidates for the presence of potentially useful pharmaceutical agents. It will be understood that the pharmaceutical agents to be screened could also be derived or synthesized from chemical compositions or man-made compounds. Thus, it is understood that the candidate substance identified by the present invention may be polypeptide, polynucleotide, small molecule inhibitors or any other compounds that may be designed through rational drug design starting from known inhibitors of Src family tyrosine kinases, nephropathy or kidney other disease.

Detail Description Paragraph - DETX (132):

[0172] The target may be either free in solution, fixed to support, expressed in or on the surface of a cell. Either the target or the compound may be labeled, thereby permitting determining of binding. In another embodiment, the assay may measure the inhibition of binding of a target to a natural or artificial substrate or binding partner (such as Nef and a member of the Src tyrosine kinase family). Competitive binding assays can be performed in which one of the agents (Nef, for example) is labeled. Usually, the target will be the labeled species, decreasing the chance that the labeling will interfere with the binding moiety's function. One may measure the amount of free label versus bound label to determine, binding or inhibition of binding.

Detail Description Paragraph - DETX (141):

[0181] The present invention particularly contemplates the use of various animal models. Here, transgenic mice are contemplated and provide a specific model for HIVAN in a whole animal system. The generation of these animals has been described elsewhere in this document. These models can, therefore be used not only screen for inhibitors of the Nef/Src interaction but also to track the progression of nephropathic disease.

Detail Description Paragraph - DETX (145):

[0185] In the sections above, the present invention describes various novel compositions for the inhibition of the Nef/Src interaction, also described are assays for identifying additional composition. It is contemplated that therapeutic compositions of the present invention will be useful in the intervention of various disease states such as for example, HIVAN, AIDS dementia; HIV-induced anemia; HIV-induced lymphoma; HIV-induced myopathy; HIV-induced cardiomyopathy; and primary HIV-induced disease progression and any other disorders mediated through the interaction of HIV-1 Nef with Src family tyrosine kinases. Such agents may be used either alone or in combination with other therapeutic agents presently being used to control the deleterious effects of HIV-1 infection. In order to be used in such therapeutic indications, it will be preferable to prepare the compositions of the invention

in pharmaceutically acceptable formats.

Detail Description Paragraph - DETX (146):
[0186] Also, it should be understood that it may well be that purified compositions that inhibit the interaction of Nef with a member of the Src family of tyrosine kinases may be routinely prepared into pharmaceutically acceptable forms of the proteins once they are isolated from the media and/or cellular compositions described above. Generally, this will entail preparing compositions that are essentially free of pyrogens, as well as other impurities that could be harmful to humans or animals.

Detail Description Paragraph - DETX (216):
[0241] Previous studies have shown that Nef binds to SH3 domains of Src kinase family members and promotes their transforming activities (reviewed in refs. 19). The expression of Src family members was examined by RT-PCR and immunoprecipitation. These investigations revealed that that Hck, Lyn, Lck, and Src were expressed in podocytes transduced with Nef or vector. To explore a possible role for these kinases in Nef-induced loss of contact inhibition in podocytes, the specific activity of several Src tyrosine kinases was determined. Src and Hck were immunoprecipitated from podocytes transduced with Nef or vector, and then incubated with [.gamma.-32P]-ATP and enolase. As shown in FIG. 13, the specific activity of the Src tyrosine kinase was increased 2.3-fold in cells expressing Nef. The expression level of Src protein showed no change with Nef by western blotting. In contrast, both the expression and activity level of Hck increased in Nef-expressing cells. These data suggest that Nef induces Src and Hck activities in podocytes.

Claims Text - CLTX (2):
1. A method of inhibiting kidney cell dedifferentiation, comprising inhibiting the interaction of Nef with a Src family tyrosine kinase SH3 domain of a polypeptide of said cell.

Claims Text - CLTX (6):
5. The method of claim 1, wherein said inhibiting the interaction of Nef with a SH3 domain of a Src family tyrosine kinase comprises reducing the expression of Nef in said cell.

Claims Text - CLTX (7):
6. The method of claim 1, wherein said inhibiting the interaction of Nef with a SH3 domain of a Src family tyrosine kinase comprises contacting said Nef with an agent that binds to and/or inactivates said Nef.

Claims Text - CLTX (46):
45. A method of treating a subject, comprising inhibiting the interaction of Nef with a SH3 domain of a Src family tyrosine kinase, wherein said subject has a disease associated with HIV-1 infection.

PGPUB-DOCUMENT-NUMBER: 20030225098

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20030225098 A1

TITLE: Kinase inhibitors

PUBLICATION-DATE: December 4, 2003

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Hirst, Gavin C.	Princeton	MA	US	
Arnold, Lee D.	Niantic	CT	US	
Burchat, Andrew	Shrewsbury	MA	US	
Wishart, Neil	Holden	MA	US	
Calderwood, David	Framingham	MA	US	
Wada, Carol K.	Gurnee	IL	US	
Michaelides, Michael R.	Libertyville	IL	US	
Ji, Zhiqin	Libertyville	IL	US	
Muckey, Melanie	Trevor	WI	US	

APPL-NO: 10/ 394965

DATE FILED: March 21, 2003

RELATED-US-APPL-DATA:

non-provisional-of-provisional 60366422 20020321 US

US-CL-CURRENT: 514/252.02, 514/255.05 , 514/260.1 , 514/262.1 , 514/263.2 , 514/263.22 , 514/81 , 544/238 , 544/244 , 544/254 , 544/262 , 544/277 , 544/280

ABSTRACT:

The present application is directed to pyrazolopyrirnidine and furopyrimidine analogs of the formula (I) 1
wherein the substituents are as defined herein, which are useful as kinase inhibitors.

---------- KWIC ---------

Summary of Invention Paragraph - BSTX (10):
[0009] As previously stated, recent evidence suggests that VEGF plays a role in the stimulation of both normal and pathological angiogenesis (Jakeman et al., Endocrinology 133: 848-859, 1993; Kolch et al., Breast Cancer Research and Treatment 36: 139-155, 1995; Ferrara et al., Endocrine Reviews 18(1); 4-25, 1997; Ferrara et al., Regulation of Angiogenesis (ed. L. D. Goldberg and E. M. Rosen), 209-232, 1997). In addition, VEGF has been implicated in the control and enhancement of vascular permeability (Connolly, et al., J. Biol. Chem. 264: 20017-20024, 1989; Brown et al., Regulation of Angiogenesis (ed. L. D. Goldberg and E. M. Rosen), 233-269, 1997). Different forms of VEGF arising from alternative splicing of mRNA have been reported, including the four species described by Ferrara et al. (J. Cell. Biochem. 47:211-218, 1991). Both secreted and predominantly cell-associated species of VEGF have been identified by Ferrara et al. supra, and the protein is known to exist in the

form of disulfide linked dimers.

Summary of Invention Paragraph - BSTX (12):
[0011] Placenta growth factor (PlGF) has an amino acid sequence that exhibits significant homology to the VEGF sequence (Park et al., J. Biol. Chem. 269:25646-54, 1994; Maglione et al. Oncogene 8:925-31, 1993). As with VEGF, different species of PlGF arise from alternative splicing of mRNA, and the protein exists in dimeric form (Park et al., supra). PlGF-1 and PlGF-2 bind to Flt-1 with high affinity, and PlGF-2 also avidly binds to neuropilin-1 (Migdal et al, J. Biol. Chem. 273 (35): 22272-22278), but neither binds to FLK-1/KDR (Park et al., supra). PlGF has been reported to potentiate both the vascular permeability and mitogenic effect of VEGF on endothelial cells when VEGF is present at low concentrations (purportedly due to heterodimer formation) (Park et al., supra).

Summary of Invention Paragraph - BSTX (16):
[0015] As for VEGF, VEGF-C and VEGF-D have been claimed to induce increases in vascular permeability in vivo in a Miles assay when injected into cutaneous tissue (PCT/US97/14696; WO98/07832, Witzenbichler et al., supra). The physiological role and significance of these ligands in modulating vascular hyperpermeability and endothelial responses in tissues where they are expressed remains uncertain.

Summary of Invention Paragraph - BSTX (18):
[0017] Based upon emerging discoveries of other homologs of VEGF and VEGFRs and the precedents for ligand and receptor heterodimerization, the actions of such VEGF homologs may involve formation of VEGF ligand heterodimers, and/or heterodimerization of receptors, or binding to a yet undiscovered VEGFR (Witzenbichler et al., supra). Also, recent reports suggest neuropilin-1 (Migdal et al, supra) or VEGFR-3/Flt-4 (Witzenbichler et al., supra), or receptors other than KDR/VEGFR-2 may be involved in the induction of vascular permeability (Stacker, S. A., Vitali, A., Domagala, T., Nice, E., and Wilks, A. F., AAngiogenesis and Cancer.congruent.Conference, Amer. Assoc. Cancer Res., Jan. 1998, Orlando, Fla.; Williams, Diabetelogia 40: S 18-120 (1997)). Until now, no direct evidence for the essential role of KDR in VEGF-mediated vascular hyperpermeability has been disclosed.

Summary of Invention Paragraph - BSTX (22):
[0021] More recently, attempts have been made to identify small molecules which act as tyrosine kinase inhibitors. For example, bis monocyclic, bicyclic or heterocyclic aryl compounds (PCT WO 92/20642) and vinylene-azaindole derivatives (PCT WO 94/14808) have been described generally as tyrosine kinase inhibitors. Styryl compounds (U.S. Pat. No. 5,217,999), styryl-substituted pyridyl compounds (U.S. Pat. No. 5,302,606), certain quinazoline derivatives (EP Application No. 0 566 266 A1; Expert Opin. Ther. Pat. (1998), 8(4): 475478), selenoindoles and selenides (PCT WO 94/03427), tricyclic polyhydroxylic compounds (PCT WO 92/21660) and benzylphosphonic acid compounds (PCT WO 91/15495) have been described as compounds for use as tyrosine kinase inhibitors for use in the treatment of cancer. Anilinocinnolines (PCT WO97/34876) and quinazoline derivative compounds (PCT WO97/22596; PCT WO97/42187) have been described as inhibitors of angiogenesis and vascular permeability.

Summary of Invention Paragraph - BSTX (23):
[0022] In addition, attempts have been made to identify small molecules which act as serine/threonine kinase inhibitors. For example, bis(indolylmaleimide) compounds have been described as inhibiting particular PKC serine/threonine kinase isoforms whose signal transducing function is associated with altered vascular permeability in VEGF-related diseases (PCT

WO97/40830; PCT WO97/40831).

Summary of Invention Paragraph - BSTX (28):
[0027] The identification of effective small compounds which specifically inhibit signal transduction and cellular proliferation by modulating the activity of receptor and non-receptor tyrosine and serine/threonine kinases to regulate and modulate abnormal or inappropriate cell proliferation, differentiation, or metabolism is therefore desirable. In particular, the identification of methods and compounds that specifically inhibit the function of a tyrosine kinase which is essential for antiogenic processes or the formation of vascular hyperpermeability leading to edema, ascites, effusions, exudates, and macromolecular extravasation and matrix deposition as well as associated disorders would be beneficial.

Summary of Invention Paragraph - BSTX (169):
[0167] a method of treating a condition in a patient comprising administering a therapeutically effective amount of a compound of formula (I) or a physiologically acceptable salt, prodrug or biologically active metabolites thereof to said patient, wherein said condition is an ocular condition, a cardiovascular condition, a cancer, Crow-Fukase (POEMS) syndrome, a diabetic condition, sickle cell anaemia, chronic inflammation, systemic lupus, glomerulonephritis, synovitis, inflammatory bowel disease, Crohn's disease, glomerulonephritis, rheumatoid arthritis, osteoarthritis, multiple sclerosis, graft rejection, Lyme disease, sepsis, von Hippel Lindau disease, pemphigoid, psoriasis, Paget's disease, polycystic kidney disease, fibrosis, sarcoidosis, cirrhosis, thyroiditis, hyperviscosity syndrome, Osler-Weber-Rendu disease, chronic occlusive pulmonary disease, asthma or edema following burns, trauma, radiation, stroke, hypoxia, ischemia, ovarian hyperstimulation syndrome, preeclampsia, menometrorrhagia, endometriosis, pulmonary hypertension, infantile hemangioma, or infection by Herpes simplex, Herpes Zoster, human immunodeficiency virus, parapoxvirus, protozoa or toxoplasmosis;

Summary of Invention Paragraph - BSTX (170):
[0168] a method wherein the ocular condition is ocular or macular edema, ocular neovascular disease, scleritis, radial keratotomy, uveitis, vitritis, myopia, optic pits, chronic retinal detachment, post-laser treatment complications, conjunctivitis, Stargardt's disease, Eales disease, retinopathy or macular degeneration;

Summary of Invention Paragraph - BSTX (185):
[0182] Inhibitors of kinases involved in mediating or maintaining these disease states represent novel therapies for these disorders. Examples of such kinases include, but are not limited to: (1) inhibition of c-Src (Brickell, Critical Reviews in Oncogenesis, 3:401-406 (1992); Courtneidge, Seminars in Cancer Biology, 5:236-246 (1994), raf (Powis, Pharmacology & Therapeutics, 62:57-95 (1994)) and the cyclin-dependent kinases (CDKs) 1, 2 and 4 in cancer (Pines, Current Opinion in Cell Biology, 4:144-148 (1992); Lees, Current Opinion in Cell Biology, 7:773-780 (1995); Hunter and Pines, Cell, 79:573-582 (1994)), (2) inhibition of CDK2 or PDGF-R kinase in restenosis (Buchdunger et al., Proceedings of the National Academy of Science USA, 92:2258-2262 (1995)), (3) inhibition of CDK5 and GSK3 kinases in Alzheimers (Hosoi et al., Journal of Biochemistry (Tokyo), 117:741-749 (1995); Aplin et al., Journal of Neurochemistry, 67:699-707 (1996), (4) inhibition of c-Src kinase in osteoporosis (Tanaka et al., Nature, 383:528-531 (1996), (5) inhibition of GSK-3 kinase in type-2 diabetes (Borthwick et al., Biochemical & Biophysical Research Communications, 210:738-745 (1995), (6) inhibition of the p38 kinase in inflammation (Badger et al., The Journal of Pharmacology and Experimental Therapeutics, 279:1453-1461 (1996)), (7) inhibition of VEGF-R 1-3 and TIE-1 and -2 kinases in diseases which involve angiogenesis (Shawver et al., Drug

Discovery Today, 2:50-63 (1997)), (8) inhibition of UL97 kinase in viral infections (He et al., Journal of Virology, 71:405-411 (1997)), (9) inhibition of CSF-1R kinase in bone and hematopoetic diseases (Myers et al., Bioorganic & Medicinal Chemistry Letters, 7:421-424 (1997), and (10) inhibition of Lck kinase in autoimmune diseases and transplant rejection (Myers et al., Bioorganic & Medicinal Chemistry Letters, 7:417-420 (1997)).

Summary of Invention Paragraph - BSTX (265):

[0262] Further, some of these compounds can be used as active agents against burns, chronic lung disease, stroke, polyps, anaphylaxis, chronic and allergic inflammation, delayed-type hypersensitivity, ovarian hyperstimulation syndrome, brain tumor-associated cerebral edema, high-altitude, trauma or hypoxia induced cerebral or pulmonary edema, ocular and macular edema, ascites, glomerulonephritis and other diseases where vascular hyperpenneability, effusions, exudates, protein extravasation, or edema is a manifestation of the disease. The compounds will also be useful in treating disorders in which protein extravasation leads to the deposition of fibrin and extracellular matrix, promoting stromal proliferation (e.g. keloid, fibrosis, cirrhosis and carpal tunnel syndrome). Increased VEGF production potentiates inflammatory processes such as monocyte recruitment and activation. The compounds of this invention will also be useful in treating inflammatory disorders such as inflammatory bowel disease (IBD) and Crohn's disease.

Summary of Invention Paragraph - BSTX (266):

[0263] VEGF's are unique in that they are the only angiogenic growth factors known to contribute to vascular hyperpermeability and the formation of edema. Indeed, vascular hyperpermeability and edema that is associated with the expression or administration of many other growth factors appears to be mediated via VEGF production. Inflammatory cytokines stimulate VEGF production. Hypoxia results in a marked upregulation of VEGF in numerous tissues, hence situations involving infarct, occlusion, ischeria, anemia, or circulatory impairment typically invoke VEGFJVPF mediated responses. Vascular hyperpermeability, associated edema, altered transendothelial exchange and macromolecular extravasation, which is often accompanied by diapedesis, can result in excessive matrix deposition, aberrant stromal proliferation, fibrosis, etc. Hence, VEGF-mediated hyperpermeability can significantly contribute to disorders with these etiologic features.

Summary of Invention Paragraph - BSTX (272):

[0269] The compounds of this invention have inhibitory activity against protein kinases. That is, these compounds modulate signal transduction by protein kinases. Compounds of this invention inhibit protein kinases from serine/threonine and tyrosine kinase classes. In particular, these compounds selectively inhibit the activity of the KDR/FLK-1/VEGFR-2 tyrosine kinases. Certain compounds of this invention also inhibit the activity of additional tyrosine kinases such as Flt-1/VEGFR-1, Flt-4/VEGFR-3, Tie-1, Tie-2, FGFR, PDGFR, IGF-1R, c-Met, Src-subfamily kinases such as Lck, hck, fgr, Src, fyn, yes, etc. Additionally, some compounds of this invention significantly inhibit serine/threonine kinases such as PKC, MAP kinases, erk, CDKs, Plk-1, or Raf-1 which play an essential role in cell proliferation and cell-cycle progression. The potency and specificity of the generic compounds of this invention towards a particular protein kinase can often be altered and optimized by variations in the nature, number and arrangement of the substituents (i.e., R.sub.1, R.sub.2, R.sub.3, A and ring 1) and conformational restrictions. In addition the metabolites of certain compounds may also possess significant protein kinase inhibitory activity.

Summary of Invention Paragraph - BSTX (273):

[0270] The compounds of this invention, when administered to individuals in

need of such compounds, inhibit vascular hyperpermeability and the formation of edema in these individuals. These compounds act, it is believed, by inhibiting the activity of KDR tyrosine kinase which is involved in the process of vascular hyperpermeability and edema formation. The KDR tyrosine kinase may also be referred to as FLK-1 tyrosine kinase, NYK tyrosine kinase or VEGFR-2 tyrosine kinase. KDR tyrosine kinase is activated when vascular endothelial cell growth factor (VEGF) or another activating ligand (such as VEGF-C, VEGF-D, VEGF-E or HIV Tat protein) binds to a KDR tyrosine kinase receptor which lies on the surface of vascular endothelial cells. Following such KDR tyrosine kinase activation, hyperpermeability of the blood vessels occurs and fluid moves from the blood stream past the blood vessel walls into the interstitial spaces, thereby forming an area of edema. Diapedesis also often accompanies this response. Similarly, excessive vascular hyperpermeability can disrupt normal molecular exchange across the endothelium in critical tissues and organs (e.g., lung and kidney), thereby causing macromolecular extravasation and deposition. Following this acute response to KDR stimulation which is believed to facilitate the subsequent angiogenic process, prolonged KDR tyrosine kinase stimulation results in the proliferation and chemotaxis of vascular endothelial cells and formation of new vessels. By inhibiting KDR tyrosine kinase activity, either by blocking the production of the activating ligand, by blocking the activating ligand binding to the KDR tyrosine kinase receptor, by preventing receptor dimerization and transphosphorylation, by inhibiting the enzyme activity of the KDR tyrosine kinase (inhibiting the phosphorylation function of the enzyme) or by some other mechanism that interrupts its downstream signaling (D. Mukhopedhyay et al., Cancer Res. 58:1278-1284 (1998) and references therein), hyperpermeability, as well as associated extravasation, subsequent edema formation and matrix deposition, and angiogenic responses, may be inhibited and minimized.

Summary of Invention Paragraph - BSTX (279):

[0276] The method of the present invention is useful in the treatment of protein kinase-mediated conditions, such as any of the conditions described above. In one embodiment, the protein kinase-mediated condition is characterized by undesired angiogenesis, edema, or stromal deposition. For example, the condition can be one or more ulcers, such as ulcers caused by bacterial or fungal infections, Mooren ulcers and ulcerative colitis. The condition can also be due to a microbial infection, such as Lyme disease, sepsis, septic shock or infections by Herpes simplex, Herpes Zoster, human immunodeficincy virus, protozoa, toxoplasmosis or parapoxvirus; an angiogenic disorders, such as von Hippel Lindau disease, polycystic kidney disease, pemphigoid, Paget's disease and psoriasis; a reproductive condition, such as endometriosis, ovarian hyperstimulation syndrome, preeclampsia or menometrorrhagia; a fibrotic and edemic condition, such as sarcoidosis, fibrosis, cirrhosis, thyroiditis, hyperviscosity syndrome systemic, Osler-Weber-Rendu disease, chronic occlusive pulmonary disease, asthma, and edema following burns, trauma, radiation, stroke, hypoxia or ischemia; or an inflammatory/immunologic condition, such as systemic lupus, chronic inflammation, glomerulonephritis, synovitis, inflammatory bowel disease, Crohn's disease, rheumatoid arthritis, osteoarthritis, multiple sclerosis and graft rejection. Suitable protein kinase-mediated conditions also include sickle cell anaemia, osteoporosis, osteopetrosis, tumor-induced hypercalcernia and bone metastases. Additional protein kinase-mediated conditions which can be treated by the method of the present invention include ocular conditions such as ocular and macular edema, ocular neovascular disease, scleritis, radial keratotomy, uveitis, vitritis, myopia, optic pits, chronic retinal detachment, post-laser complications, conjunctivitis, Stargardt's disease and Eales disease, in addition to retinopathy and macular degeneration.

Summary of Invention Paragraph - BSTX (285):

[0282] In many pathological conditions (for example, solid primary tumors and metastases, Kaposi's sarcoma, rheumatoid arthritis, blindness due to inappropriate ocular neovascularization, psoriasis and atherosclerosis) disease progression is contingent upon persistent angiogenesis. Polypeptide growth factors often produced by the disease tissue or associated inflammatory cells, and their corresponding endothelial cell specific receptor tyrosine kinases (e.g., KDR/VEGFR-2, Flt-1/VEGFR-1, Tie-2/Tek and Tie) are essential for the stimulation of endothelial cell growth, migration, organization, differentiation and the establishment of the requisite new functional vasculature. As a result of the vascular permeability factor activity of VEGF in mediating vascular hyperpermeability, VEGF-stimulation of a VEGFR kinase is also believed to play an important role in the formation of tumor ascites, cerebral and pulmonary edema, pleural and pericardial effusions, delayed-type hypersensitivity reactions, tissue edema and organ dysfunction following trauma, bums, ischemia, diabetic complications, endometriosis, adult respiratory distress syndrome (ARDS), post-cardiopulmonary bypass-related hypotension and hyperpermeability, and ocular edema leading to glaucoma or blindness due to inappropriate neovascularization. In addition to VEGF, recently identified VEGF-C and VEGF-D, and virally-encoded VEGF-E or HIV-Tat protein can also cause a vascular hyperpermeability response through the stimulation of a VEGFR kinase. KDR/VEGFR-2 and/or Tie-2 are expressed also in a select population of hematopoietic stem cells. Certain members of this population are pluripotent in nature and can be stimulated with growth factors to differentiate into endothelial cells and participate in vasculogenetic angiogenic processes. For this reason these have been called Endothelial Progenitor Cells (EPCs) (J. Clin. Investig. 103: 1231-1236 (1999)). In some progenitors, Tie-2 may play a role in their recruitment, adhesion, regulation and differentiation (Blood, 4317-4326 (1997)). Certain agents according to formula (I) capable of blocking the kinase activity of endothelial cell specific kinases could therefore inhibit disease progression involving these situations.

Summary of Invention Paragraph - BSTX (287):

[0284] The compounds of formula (I) or a salt thereof or pharmaceutical compositions containing a therapeutically effective amount thereof may be used in the treatment of protein kinase-mediated conditions, such as benign and neoplastic proliferative diseases and disorders of the immune system, as described above. For example, such diseases include autoimmune diseases, such as rheumatoid arthritis, thyroiditis, type 1 diabetes, multiple sclerosis, sarcoidosis, inflammatory bowel disease, Crohn's disease, myasthenia gravis and systemic lupus erythematosus; psoriasis, organ transplant rejection (eg. kidney rejection, graft versus host disease), benign and neoplastic proliferative diseases, human cancers such as lung, breast, stomach, bladder, colon, pancreas, ovarian, prostate and rectal cancer and hematopoietic malignancies (leukemia and lymphoma), and diseases involving inappropriate vascularization for example diabetic retinopathy, retinopathy of prematurity, choroidal neovascularization due to age-related macular degeneration, and infantile hemangiomas in human beings. In addition, such inhibitors may be useful in the treatment of disorders involving VEGF mediated edema, ascites, effusions, and exudates, including for example macular edema, cerebral edema, acute lung injury and adult respiratory distress syndrome (ARDS).

Summary of Invention Paragraph - BSTX (294):

[0291] The compounds of this invention can be administered to a human patient by themselves or in pharmaceutical compositions where they are mixed with suitable carriers or excipient(s) at doses to treat or ameliorate vascular hyperpermeability, edema and associated disorders. Mixtures of these compounds can also be administered to the patient as a simple mixture or in suitable formulated pharmaceutical compositions. A therapeutically effective dose

further refers to that amount of the compound or compounds sufficient to result in the prevention or attenuation of inappropriate neovascularization, progression of hyperproliferative disorders, edema, VEGF-associated hyperpermeability and/or VEGF-related hypotension. Techniques for formulation and administration of the compounds of the instant application may be found in "Remington's Pharmaceutical Sciences," Mack Publishing Co., Easton, Pa., latest edition.

Summary of Invention Paragraph - BSTX (297):
[0294] Alternatively, one may administer the compound in a local rather than a systenic manner, for example, via injection of the compound directly into an edematous site, often in a depot or sustained release formulation.

Summary of Invention Paragraph - BSTX (337):
[0334] In the compositions of the present invention the active compound may, if desired, be associated with other compatible pharmacologically active ingredients. For example, the compounds of this invention can be administered in combination with one or more additional pharmaceutical agents that inhibit or prevent the production of VEGF or angiopoietins, attenuate intracellular responses to VEGF or angiopoietins, block intracellular signal transduction, inhibit vascular hyperpermeability, reduce inflammation, or inhibit or prevent the formation of edema or neovascularization. The compounds of the invention can be administered prior to, subsequent to or simultaneously with the additional pharmaceutical agent, whichever course of administration is appropriate. The additional pharmaceutical agents include but are not limited to anti-edemic steroids, NSAIDS, ras inhibitors, anti-TNF agents, anti-IL1 agents, antihistamines, PAF-antagonists, COX-1 inhibitors, COX-2 inhibitors, NO synthase inhibitors, Akt/PTB inhibitors, IGF-IR inhibitors, PKC inhibitors and PI3 kinase inhibitors. The compounds of the invention and the additional pharmaceutical agents act either additively or synergistically. Thus, the administration of such a combination of substances that inhibit angiogenesis, vascular hyperpermeability and/or inhibit the formation of edema can provide greater relief from the deletrious effects of a hyperproliferative disorder, angiogenesis, vascular hyperpermeability or edema than the administration of either substance alone. In the treatment of malignant disorders combinations with antiproliferative or cytotoxic chemotherapies or radiation are anticipated.

Summary of Invention Paragraph - BSTX (433):
[0430] In vivo Uterine Edema Model

Summary of Invention Paragraph - BSTX (434):
[0431] This assay measures the capacity of compounds to inhibit the acute increase in uterine weight in mice which occurs in the first few hours following estrogen stimulation. This early onset of uterine weight increase is known to be due to edema caused by increased permeability of uterine vasculature. Cullinan-Bove and Koss (Endocrinology (1993), 133:829-837) demonstrated a close temporal relationship of estrogen-stimulated uterine edema with increased expression of VEGF mRNA in the uterus. These results have been confirmed by the use of neutralizing monoclonal antibody to VEGF which significantly reduced the acute increase in uterine weight following estrogen stimulation (WO 97/42187). Hence, this system can serve as a model for in vivo inhibition of VEGF signalling and the associated hyperpermeability and edema.

Claims Text - CLTX (24):
23. A method of treating a condition in a patient comprising administering a therapeutically effective amount of a compound of claim 1 or a physiologically acceptable salt, prodrug or biologically active metabolites thereof to said patient, wherein said condition is an ocular condition, a

cardiovascular condition, a cancer, Crow-Fukase (POEMS) syndrome, a diabetic condition, sickle cell anaemia, chronic inflammation, systemic lupus, glomerulonephritis, synovitis, inflammatory bowel disease, Crohn's disease, glomerulonephritis, rheumatoid arthritis, osteoarthritis, multiple sclerosis, graft rejection, Lyme disease, sepsis, von Hippel Lindau disease, pemphigoid, psoriasis, Paget's disease, polycystic kidney disease, fibrosis, sarcoidosis, cirrhosis, thyroiditis, hyperviscosity syndrome, Osler-Weber-Rendu disease, chronic occlusive pulmonary disease, asthma or edema following burns, trauma, radiation, stroke, hypoxia, ischemia, ovarian hyperstimulation syndrome, preeclampsia, menometrorrhagia, endometriosis, pulmonary hypertension, infantile hemangioma, or infection by Herpes simplex, Herpes Zoster, human immunodeficiency virus, parapoxvirus, protozoa or toxoplasmosis.

Claims Text - CLTX (25):

24. The method of claim 23 wherein the ocular condition is ocular or macular edema, ocular neovascular disease, scleritis, radial keratotomy, uveitis, vitritis, myopia, optic pits, chronic retinal detachment, post-laser treatment complications, conjunctivitis, Stargardt's disease, Eales disease, retinopathy or macular degeneration.

US-PAT-NO: 6764833

DOCUMENT-IDENTIFIER: US 6764833 B1

TITLE: Mutated Src oncogene composition and methods

DATE-ISSUED: July 20, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Yeatman; Timothy J.	Tampa	FL	N/A	N/A
Irby; Rosalyn B.	Tampa	FL	N/A	N/A

APPL-NO: 09/ 444711

DATE FILED: November 24, 1999

US-CL-CURRENT: 435/69.1, 435/320.1 , 435/325 , 530/300 , 530/350 , 536/1.11 , 536/18.7 , 536/22.1 , 536/23.1 , 536/23.2 , 536/23.5

ABSTRACT:

The present invention provides a mutant oligonucleotide composition encoding a cellular c-Src tyrosine kinase oncogene. Methods for isolating, expressing and characterizing recombinant Src mutant polypeptide are also provided. The invention further relates to methods for utilizing such oligonucleotides, polypeptides, agonists and antagonists for applications, which relate to research, diagnostics, and clinical arts. More specifically, this invention provides methods of diagnosing, treating, immunizing, and creating transgenic animals based on use of such mutant Src.

20 Claims, 14 Drawing figures

Exemplary Claim Number: 1

Number of Drawing Sheets: 4

---------- KWIC ---------

Brief Summary Text - BSTX (8):

The non receptor tyrosine kinase c-Src consists of an SH3, SH2 and tyrosine kinase domain. c-Src appears to be the most important to the normal function of osteoclasts, as determined from studies of Src-knock-out mice (see for example U.S. Pat. No. 5,541,109). The catalytic activity of c-Src and other nonreceptor tyrosine kinases is inhibited by the intramolecular association of their intrinsic SH2 domain to the carboxy-terminal tail upon phosphorylation of Tyr (position 530, avian position 527). Protein tyrosine phosphorylation is believed to be an important regulatory event in cell growth and differentiation. Phosphorylation on tyrosine can either decrease or increase the enzymatic activity of substrate proteins. Tyrosine phosphorylated sequences associate with Src homology 2 (SH2) domains, and thus tyrosine phosphorylation also serves to regulate protein/protein interactions. Many protein tyrosine kinases have been described to date: several are the receptors for peptide growth factors; others are expressed in the cytoplasm and nucleus. Tyrosine kinases can be of the receptor type (having extracellular, transmembrane and intracellular domains) or the non-receptor type (being wholly

intracellular). There are 19 known families of receptor tyrosine kinases including the Her family (EGFR, Her 2, Her 3, Her 4), the insulin receptor family (insulin receptor, IGF-1R, insulin-related receptor), the PDGF receptor family (PDGF-R alpha and beta, CSF-1R, Kit, Flk2), the Flk family (Flk-1, Flt-1, Flk-4), the FGF-receptor family (FGF-Rs 1 through 4), the Met family (Met, Ron), etc. There are 11 known families of non-receptor type tyrosine kinases including the Src family (Src, Yes, Fyn, Lyn, Lck, Blk, Hck, Fgr, Yrk), Abl family (Abl, Arg), Zap 70 family (Zap 70, Syk) and Jak family (Jak 1, Jak 2, Tyk 2, Jak 3). Many of these tyrosine kinases have been found to be involved in cellular signaling pathways leading to pathogenic conditions such as cancer, psoriasis, hyperimmune response, etc. Other roles for tyrosine kinases include cellular responses to a variety of extracellular signals, such as those arising from growth factors and cell-cell interactions, as well as in differentiating developmental processes in both vertebrates and invertebrates.

Brief Summary Text - BSTX (14):

This invention also provides a method of screening agonist and antagonist compounds for the treatment of mutant Src associated or caused diseases. A method of treating a cancer is provided by administering to cancerous cells exhibiting a c-Src mutation at SRC 531 an effective amount of a compound capable of inhibiting the excess kinase activity resulting from the c-Src mutation or capable of inhibiting expression of the c-Src mutant gene (SEQ ID NO. 3). Preferred compounds of the invention comprise an antisense oligonucleotide, or a preparation of antibodies, or other molecules which specifically bind to c-Src SRC 531 mutant (SEQ ID NO. 3).

Detailed Description Text - DETX (21):

Antisense oligonucleotides according to the invention are perfectly suitable for the inhibition of mutant c-Src expression. In the context of this invention, the term "oligonucleotide" refers to an oligomer or polymer of ribonucleic acid (RNA) or deoxyribonucleic acid (DNA) or mimetics thereof. Thus, they represent preparations for inhibiting the over-expressed c-Src in well-calculated fashion. This offers new possibilities of being able to treat by means of gene therapy various diseases linked with the anomalous tyrosine kinase biosynthesis, particularly tumor diseases. The antisense compounds in accordance with this invention preferably comprise from about 8 to about 120 nucleobases. Particularly preferred are antisense oligonucleotides comprising from about 8 to about 30 nucleobases (i.e. from about 8 to about 30 linked nucleosides). As is known in the art, a nucleoside is a base-sugar combination. The base portion of the nucleoside is normally a heterocyclic base. The two most common classes of such heterocyclic bases are the purines and the pyrimidines. Nucleotides are nucleosides that further include a phosphate group covalently linked to the sugar portion of the nucleoside. For those nucleosides that include a pentofuranosyl sugar, the phosphate group can be linked to either the 2', 3' or 5' hydroxyl moiety of the sugar. In forming oligonucleotides, the phosphate groups covalently link adjacent nucleosides to one another to form a linear polymeric compound. In turn the respective ends of this linear polymeric structure can be further joined to form a circular structure, however, open linear structures are generally preferred. Within the oligonucleotide structure, the phosphate groups are commonly referred to as forming the internucleoside backbone of the oligonucleotide. The normal linkage or backbone of RNA and DNA is a 3' to 5' phosphodiester linkage.

Detailed Description Text - DETX (98):

A library of peptides to be tested as antagonists of pp60 c-Src mutant tyrosine kinase are synthesized according to the procedure disclosed in U.S. Pat. No. 5,532,167 to Cantley, which is incorporated herein by way of reference. Accordingly a peptide Ala-Glu-Glu-Glu-Ile-Tyr-Gly-Glu-Phe-Glu-Ala-Lys-Lys-Lys-Lys is synthesized as

an optimal substrate/antagonist for mutant Src tyrosine kinase. The kinetic studies are carried out as follows. Purified mutant kinase is immobilized on protein A beads and kinase reaction is performed in 20 .mu.l of 50 mM Tris pH 7.0 buffer containing 50 mM NaCl, 10 mM MgCl.sub.2, 10 .mu.M ATP 5 .mu.Ci [gamma-.sup.32 P]-ATP (3000 mCi/mmol, NEN) and various dilutions of the peptide. For the experiment measuring the competitive inhibition of c-Src activity by the Src motif-containing peptide, 1 .mu.M (final concentration) acid treated enolase (trans-phosphorylation) is included. After 2.5 minutes incubation, the supernatants are spotted on phosphocellulose paper, washed four times with 75 mM phosphoric acid, and radioactivity counted in a scintillation counter. For phosphorylation of enolase, the reaction is stopped by adding SDS loading buffer and the proteins are resolved on 10% SDS-PAGE gels. The Km and Vmax are calculated using a standard computer software. The peptide is a good substrate for pp60c-Src, with Km of 2 .mu.M and Vmax of 0.9 .mu.M/mg/min. The Src-substrate peptide is also an excellent competitive inhibitor of enolase phosphorylation by pp60c-Src (K.sub.50% =5 .mu.M).

Detailed Description Text - DETX (100):

This example is not limiting and one skilled in the art can select shorter oligonucleotide according to established procedures. For example, a series of methoxyethylamine 3' end-cap oligodeoxynucleotides are prepared on a Biosearch 8750 DNA synthesizer, using standard H-phosphonate chemistry on controlled pore glass. The 15 or 18-base oligodeoxynucleotides are purified via DMT-on purification on a semi-prep Dynamax C-4 300A column. A secondary DMT-off purification is then performed on the same column. The oligomers are then desalted over a Pharmacia NAP-25 column, converted to the sodium form via Biorad AC 50W-X8 (Na+) 200-400 mesh polyprep column, and then passed over another NAP-25 column. The antisense oligos and their controls, which contained the same bases but in scrambled sequence, are in a similar manner. Lyophilized oligomers used in the following experiments are dissolved in PBS (1 mM stock) and sterile filtered with Millipore 0.2 micrometer disks. The sequence used for antisense inhibitory studies on SRC gene is a 27 base region of the corresponding mRNA spanning the AUG translation initiation codon. While the present invention is not limited to such sequences, antisense oligonucleotides directed against the initiation codon region of the mRNA are one type of antisense molecule believed to effectively inhibit translation of the resulting gene product. Other effective antisense molecules can be specifically targeted against the opposite end of the mRNA.

Detailed Description Text - DETX (101):

To selectively interfere with the expression of mutated SRC gene (SEQ ID NO. 3), 5 mice are injected once with 5 .mu.g/g weight of antisense, phosphorothioated oligodeoxynucleotide prepared as above and which is complementary to the initiator AUG domain in SRC mRNA or with PBS for controls. Three weeks following the injection, liver biopsies are prepared from all of these mice. Each biopsy is frozen and then sliced into this slices and hybridized with isotope labeled SRC nucleic probes. Following 3 days of exposure to emulsion autoradiography, slides are developed to create silver grains over cells containing SRC mRNAs. Labeling and number of positive cells is decreased in liver specimens of mice treated with antisense phosphorothioated oligodeoxynucleotide demonstrating that antisense interfered with mutated SRC 531 expression. In contrast, in control mice, SRCmRNA levels per cell increased by about 20-fold. The decrease of mutated SRC 531 expression is also confirmed by Western Blot studies using antibodies obtained by methods disclosed in Example 6.10.

Detailed Description Text - DETX (113):

DNA clones for microinjection are cleaved with appropriate restriction enzymes, such as Sal1, Not1, etc., and the DNA fragments electrophoresed on 1%

agarose gels in TBE buffer (U.S. Pat. No. 5,811,633). The DNA bands are visualized by staining with ethidium bromide, excised, and placed in dialysis bags containing 0.3M sodium acetate at pH 7.0. The DNA is then electroeluted into the dialysis bags, extracted with phenol-chloroform (1:1), and precipitated by two volumes of ethanol. The DNA is redissolved in 1 ml of low salt buffer (0.2M NaCl, 20 mM Tris, pH 7.4, and 1 mM EDTA) and purified on an Elutip-D column. The column is first primed with 3 ml of high salt buffer (1M NaCl, 20 mM Tris, pH 7.4, and 1 mM EDTA) followed by washing with 5 ml of low salt buffer. The DNA solutions are passed through the column for three times to bind DNA to the column matrix. After one wash with 3 ml of low salt buffer, the DNA is eluted with 0.4 ml of high salt buffer and precipitated by two volumes of ethanol. DNA concentrations are measured by absorption at 260 nm in a UV spectrophotometer. For microinjection, DNA concentrations are adjusted to about 3 .mu.g/ml in 5 mM Tris, pH 7.4 and 0.1 mM EDTA. Other methods for purification of DNA for microinjection are also known. The purified inserts form pcSrc531RI plasmids are then microinjected into the pronuclei of fertilized (C57BL/6.times.CBA)F2 mouse embryos and surviving embryos are transferred into pseudopregnant females according to standard procedures such as disclosed in U.S. Pat. Nos. 5,877,397, 5,907,078, 5,849,993, 5,602,309, 5,387,742, which are incorporated herein by way of reference. SRC531 construct is operably linked to a suitable promoter, e.g., RSV long terminal repeat (LTR), glial fibrillary acidic protein (GFAP), or human beta-globin promoter (GF). Mice that developed from injected embryos are analyzed for the presence of transgene sequences by Southern blot analysis of mutant DNA. Transgene copy number is estimated by band intensity relative to control standards containing known quantities of cloned DNA. At 3 to 8 weeks of age, cells are isolated from these animals and assayed for the presence of transgene encoded SRC 531 mutation. All of the control non-transgenic mice tested negative for expression of SRC 531. Southern blot analysis indicates that many of these mice contain one or more copies of the transgene per somatic and/or germ cell. Some mice with high levels of Src expression developed abnormally, including edemas, head deformities, eye, axial system defects and usually these mice did not survive. Surviving transgenic mice exhibit malignant and/or benign transformation early in their life. Tumors include lymphomas, thymomas, fibrosarcomas, angiosarcomas, hemangiomas, neurofibrosarcomas, etc. These mice are useful as a model for studying SRC 531 mutants in vivo for testing, for example, drugs or SRC 531 antagonists.

US-PAT-NO: 6716870

DOCUMENT-IDENTIFIER: US 6716870 B2

TITLE: Prodrugs of 3-(pyrrol-2-ylmethylidene)-2-indolinone derivatives

DATE-ISSUED: April 6, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Moon; Malcolm Wilson	Kalamazoo	MI	N/A	N/A
Morozowich; Walter	Kalamazoo	MI	N/A	N/A
Gao; Ping	Portage	MI	N/A	N/A
Koenig; Marcel	Burlingame	CA	N/A	N/A

APPL-NO: 10/ 243942

DATE FILED: September 16, 2002

PARENT-CASE:

CROSS-REFERENCE

This application is a divisional of application Ser. No. 09/863,819, filed May 24, 2001 now U.S. Pat. No. 6,482,848, which claims priority under 35 U.S.C. 119(e) to U.S. Provisional applications Serial No. 60/207,000 filed on May 24, 2000, and 60/225,045, filed on Aug. 11, 2000, the disclosures of which are incorporated herein by reference in their entirety.

US-CL-CURRENT: 514/418, 548/467 , 548/468 , 548/486

ABSTRACT:

The present invention is directed to prodrugs of certain 3-(pyrrol-2-yl-methylidene)-2-indolinone derivatives that modulate the activity of protein kinases ("PKs"). Pharmaceutical compositions comprising these compounds, methods of treating diseases related to abnormal PK activity utilizing pharmaceutical compositions comprising these compounds and methods of preparing them are also disclosed.

23 Claims, 0 Drawing figures

Exemplary Claim Number: 1

--------- KWIC ---------

Detailed Description Text - DETX (162):
This assay is used to screen for inhibitors of the tyrosine kinase Src.

Detailed Description Text - DETX (182):
Vascular Permeability Assay

Detailed Description Text - DETX (183):
Increased vascular permeability in tumor-dependent angiogenesis is due to a loosening of gap junctions in response to vascular endothelial growth factor

(VEGF). The Miles assay for vascular permeability (Miles and Miles, J. Physiol. 118: 228-257 (1952)) has been adapted to athymic mice in order to evaluate the ability of the compounds of the present invention to inhibit VEGF-induced vascular permeability in vivo.

US-PAT-NO: 6713474

DOCUMENT-IDENTIFIER: US 6713474 B2

TITLE: Pyrrolopyrimidines as therapeutic agents

DATE-ISSUED: March 30, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Hirst; Gavin C.	Malborough	MA	N/A	N/A
Calderwood; David	Framingham	MA	N/A	N/A
Munschauer; Rainer	Neustadt	N/A	N/A	DE
Arnold; Lee D.	Westborough	MA	N/A	N/A
Johnston; David N.	Nottingham	N/A	N/A	GB
Rafferty; Paul	Nottingham	N/A	N/A	GB

APPL-NO: 09/ 537167

DATE FILED: March 29, 2000

PARENT-CASE:

RELATED APPLICATIONS

This application is a Continuation-in-Part of International Application No.: PCT/US00/21560, filed Sep. 17, 1999, which claims the benefit of U.S. Provisional Application Nos. 60/100,832, filed Sep. 18, 1998; 60/100,833, filed Sep. 18, 1998; 60/100,834, filed Sep. 18, 1998, and 60/100,946, filed Sep. 18, 1998. The teachings of each of these referenced applications are expressly incorporated herein by reference in their entirety.

US-CL-CURRENT: 514/218, 514/228.5 , 514/234.2 , 514/252.16 , 514/252.18 , 514/252.19 , 514/252.2 , 514/265.1 , 540/575 , 544/117 , 544/230 , 544/280 , 544/61

ABSTRACT:

Chemical compounds having structural formula I ##STR1##

and physiologically acceptable salts and metabolites thereof, are inhibitors of serine/threonine and tyrosine kinase activity. Several of the kinases, whose activity is inhibited by these chemical compounds, are involved in immunologic, hyperproliferative, or angiogenic processes. Thus, these chemical compounds can ameliorate disease states where angiogenesis or endothelial cell hyperproliferation is a factor. These compounds can be used to treat cancer and hyper proliferative disorders, rheumatiod arthritis, disorders of the immune system, trasplant rejections and imflammatory disorders.

70 Claims, 0 Drawing figures

Exemplary Claim Number: 1

---------- KWIC ---------

Brief Summary Text - BSTX (10):

As previously stated, recent evidence suggests that VEGF plays a role in the stimulation of both normal and pathological angiogenesis (Jakeman et al., Endocrinology 133: 848-859, 1993; Kolch et al., Breast Cancer Research and Treatment 36: 139-155, 1995; Ferrara et al., Endocrine Reviews 18(1); 4-25, 1997; Ferrara et al., Regulation of Angiogenesis (ed. L. D. Goldberg and E. M. Rosen), 209-232, 1997). In addition, VEGF has been implicated in the control and enhancement of vascular permeability (Connolly, et al., J. Biol. Chem. 264: 20017-20024, 1989; Brown et al., Regulation of Angiogenesis (ed. L. D. Goldberg and E. M. Rosen), 233-269, 1997). Different forms of VEGF arising from alternative splicing of mRNA have been reported, including the four species described by Ferrara et al. (J. Cell. Biochem. 47:211-218, 1991). Both secreted and predominantly cell-associated species of VEGF have been identified by Ferrara et al. supra, and the protein is known to exist in the form of disulfide linked dimers.

Brief Summary Text - BSTX (12):

Placenta growth factor (PlGF) has an amino acid sequence that exhibits significant homology to the VEGF sequence (Park et al., J. Biol. Chem. 269:25646-54, 1994; Maglione et al. Oncogene 8:925-31, 1993). As with VEGF, different species of PlGF arise from alternative splicing of mRNA, and the protein exists in dimeric form (Park et al., supra). PlGF-1 and PlGF-2 bind to Flt-1 with high affinity, and PlGF-2 also avidly binds to neuropilin-1 (Migdal et al, J. Biol. Chem. 273 (35): 22272-22278), but neither binds to FLK-1/KDR (Park et al., supra). PlGF has been reported to potentiate both the vascular permeability and mitogenic effect of VEGF on endothelial cells when VEGF is present at low concentrations (purportedly due to heterodimer formation) (Park et al., supra).

Brief Summary Text - BSTX (16):

As for VEGF, VEGF-C and VEGF-D have been claimed to induce increases in vascular permeability in vivo in a Miles assay when injected into cutaneous tissue (PCT/US97/14696; WO98/07832, Witzenbichler et al., supra). The physiological role and significance of these ligands in modulating vascular hyperpermeability and endothelial responses in tissues where they are expressed remains uncertain.

Brief Summary Text - BSTX (18):

Based upon emerging discoveries of other homologs of VEGF and VEGFRs and the precedents for ligand and receptor heterodimerization, the actions of such VEGF homologs may involve formation of VEGF ligand heterodimers, and/or heterodimerization of receptors, or binding to a yet undiscovered VEGFR (Witzenbichler et al., supra). Also, recent reports suggest neuropilin-1 (Migdal et al, supra) or VEGFR-3/Flt-4 (Witzenbichler et al., supra), or receptors other than KDR/VEGFR-2 may be involved in the induction of vascular permeability (Stacker, S. A., Vitali, A., Domagala, T., Nice, E., and Wilks, A. F., "Angiogenesis and Cancer" Conference, Amer. Assoc. Cancer Res., January 1998, Orlando, Fla.; Williams, Diabetelogia 40: S118-120(1997)).

Brief Summary Text - BSTX (25):

More recently, attempts have been made to identify small molecules which act as tyrosine kinase inhibitors. For example, bis monocyclic, bicyclic or heterocyclic aryl compounds (PCT WO 92/20642) and vinylene-azaindole derivatives (PCT WO 94/14808) have been described generally as tyrosine kinase inhibitors. Styryl compounds (U.S. Pat. No. 5,217,999), styryl-substituted pyridyl compounds (U.S. Pat. No. 5,302,606), certain quinazoline derivatives (EP Application No. 0 566 266 A1; Expert Opin. Ther. Pat. (1998), 8(4): 475-478), selenoindoles and selenides (PCT WO 94/03427), tricyclic polyhydroxylic compounds (PCT WO 92/21660) and benzylphosphonic acid compounds (PCT WO 91/15495) have been described as compounds for use as tyrosine kinase

inhibitors for use in the treatment of cancer. Anilinocinnolines (PCT WO97/34876) and quinazoline derivative compounds (PCT WO97/22596; PCT WO97/42187) have been described as inhibitors of angiogenesis and vascular permeability.

Brief Summary Text - BSTX (26):
In addition, attempts have been made to identify small molecules which act as serine/threonine kinase inhibitors. For example, bis(indolylmaleimide) compounds have been described as inhibiting particular PKC serine/threonine kinase isoforms whose signal transducing function is associated with altered vascular permeability in VEGF-related diseases (PCT WO97/40830; PCT WO97/40831).

Brief Summary Text - BSTX (32):
Inhibitors of kinases involved in mediating or maintaining disease states represent novel therapies for these disorders. Examples of such kinases include, but are not limited to: (1) inhibition of c-Src (Brickell, Critical Reviews in Oncogenesis, 3:401-406 (1992); Courtneidge, Seminars in Cancer Biology, 5:236-246 (1994), raf (Powis, Pharmacology & Therapeutics, 62:57-95 (1994)) and the cyclin-dependent kinases (CDKs) 1, 2 and 4 in cancer (Pines, Current Opinion in Cell Biology, 4:144-148 (1992); Lees, Current Opinion in Cell Biology, 7:773-780 (1995); Hunter and Pines, Cell, 79:573-582 (1994)), (2) inhibition of CDK2 or PDGF-R kinase in restenosis (Buchdunger et al., Proceedings of the National Academy of Science USA, 92:2258-2262 (1995)), (3) inhibition of CDK5 and GSK3 kinases in Alzheimers (Hosoi et al., Journal of Biochemistry (Tokyo), 117:741-749 (1995); Aplin et al., Journal of Neurochemistry, 67:699-707 (1996), (4) inhibition of c-Src kinase in osteoporosis (Tanaka et al., Nature, 383:528-531 (1996), (5) inhibition of GSK-3 kinase in type-2 diabetes (Borthwick et al., Biochemical & Biophysical Research Communications, 210:738-745 (1995), (6) inhibition of the p38 kinase in inflammation (Badger et al., The Journal of Pharmacology and Experimental Therapeutics, 279:1453-1461 (1996)), (7) inhibition of VEGF-R 1-3 and TIE-1 and -2 kinases in diseases which involve angiogenesis (Shawver et al., Drug Discovery Today, 2:50-63 (1997)), (8) inhibition of UL97 kinase in viral infections (He et al., Journal of Virology, 71:405-411 (1997)), (9) inhibition of CSF-1R kinase in bone and hematopoetic diseases (Myers et al., Bioorganic & Medicinal Chemistry Letters, 7:421-424 (1997), and (10) inhibition of Lck kinase in autoimmune diseases and transplant rejection (Myers et al., Bioorganic & Medicinal Chemistry Letters, 7:417-420 (1997)).

Brief Summary Text - BSTX (34):
The identification of effective small compounds which specifically inhibit signal transduction and cellular proliferation by modulating the activity of receptor and non-receptor tyrosine and serine/threonine kinases to regulate and modulate abnormal or inappropriate cell proliferation, differentiation, or metabolism is therefore desirable. In particular, the identification of methods and compounds that specifically inhibit the function of a tyrosine kinase which is essential for angiogenic processes or the formation of vascular hyperpermeability leading to edema, ascites, effusions, exudates, and macromolecular extravasation and matrix deposition as well as associated disorders would be beneficial.

Brief Summary Text - BSTX (60):
The compounds of this invention are useful as inhibitors of serine/threonine and tyrosine kinases. In particular, compounds of this invention are useful as inhibitors of tyrosine kinases that are important in hyperproliferative diseases, especially in cancer and in the process of angiogenesis. For example, certain of these compounds are inhibitors of such receptor kinases as KDR, Flt-1, FGFR, PDGFR, c-Met, TIE-2 or IGF-1-R. Since certain of these

compounds are anti-angiogenic, they are important substances for inhibiting the progression of disease states where angiogenesis is an important component. Certain compounds of the invention are effective as inhbitors of such serine/threonine kinases as PKCs, erk, MAP kinases, MAP kinase kinases, MAP kinase kinase kinases, cdks, Plk-1 or Raf-1. These compounds are useful in the treatment of cancer, and hyperproliferative disorders. In addition, certain compounds are effective inhibitors of non-receptor kinases such as those of the Src (for example, Ick, blk and lyn), Tec, Csk, Jak, Map, Nik and Syk families. These compounds are useful in the treatment of cancer, hyperproliferative disorders and immunologic diseases.

Brief Summary Text - BSTX (63):

The present invention further includes the use of these compounds in pharmaceutical compositions with a pharmaceutically effective amount of the above-described compounds and a pharmaceutically acceptable carrier or excipient. These pharmaceutical compositions can be administered to individuals to slow or halt the process of angiogenesis in angiogenesis-aided diseases, or to treat edema, effusions, exudates or ascites and other conditions associated with vascular hyperpermeability. Certain pharmaceutical compositions can be administered to individuals to treat cancer and hyperproliferative disorders by inhibiting serine/threonine kinases such as cdk, Plk-1, erk, etc.

Brief Summary Text - BSTX (159):

Further, some of these compounds can be used as active agents against bums, chronic lung disease, stroke, polyps, anaphylaxis, chronic and allergic inflammation, delayed-type hypersensitivity, ovarian hyperstimulation syndrome, brain tumor-associated cerebral edema, high-altitude, trauma or hypoxia induced cerebral or pulmonary edema, ocular and macular edema, ascites, glomerulonephritis and other diseases where vascular hyperpermeability, effusions, exudates, protein extravasation, or edema is a manifestation of the disease. The compounds will also be useful in treating disorders in which protein extravasation leads to the deposition of fibrin and extracellular matrix, promoting stromal proliferation (e.g. keloid, fibrosis, cirrhosis and carpal tunnel syndrome). Increased VEGF production potentiates inflammatory processes such as monocyte recruitment and activation. The compounds of this invention will also be useful in treating inflammatory disorders such as inflammatory bowel disease (IBD) and Crohn's disease.

Brief Summary Text - BSTX (160):

VEGF's are unique in that they are the only angiogenic growth factors known to contribute to vascular hyperpermeability and the formation of edema. Indeed, vascular hyperpermeability and edema that is associated with the expression or administration of many other growth factors appears to be mediated via VEGF production. Inflammatory cytokines stimulate VEGF production. Hypoxia results in a marked upregulation of VEGF in numerous tissues, hence situations involving infarct, occlusion, ischemia, anemia, or circulatory impairment typically invoke VEGF/VPF mediated responses. Vascular hyperpermeability, associated edema, altered transendothelial exchange and macromolecular extravasation, which is often accompanied by diapedesis, can result in excessive matrix deposition, aberrant stromal proliferation, fibrosis, etc. Hence, VEGF-mediated hyperpermeability can significantly contribute to disorders with these etiologic features.

Brief Summary Text - BSTX (163):

The compounds of this invention have inhibitory activity against protein kinases. That is, these compounds modulate signal transduction by protein kinases. Compounds of this invention inhibit protein kinases from serine/threonine and tyrosine kinase classes. In particular, these compounds

selectively inhibit the activity of the KDR/FLK-1IVEGFR-2 tyrosine kinases. Certain compounds of this invention also inhibit the activity of additional tyrosine kinases such as Flt-1/VEGFR-1, Tie-2, FGFR, PDGFR, IGF-1R, c-Met, Src-subfamily kinases such as Lck, Src, fyn, yes, etc. Additionally, some compounds of this invention significantly inhibit serine/threonine kinases such as PKC, MAP kinases, erk, CDKs, Plk-1, or Raf-1 which play an essential role in cell proliferation and cell-cycle progression. The potency and specificity of the generic compounds of this invention towards a particular protein kinase can often be altered and optimized by variations in the nature, number and arrangement of the substituents (i.e., R.sub.1, R.sub.2, R.sub.3, A and ring 1) and conformational restrictions. In addition the metabolites of certain compounds may also possess significant protein kinase inhibitory activity.

Brief Summary Text - BSTX (164):

The compounds of this invention, when administered to individuals in need of such compounds, inhibit vascular hyperpermeability and the formation of edema in these individuals. These compounds act, it is believed, by inhibiting the activity of KDR tyrosine kinase which is involved in the process of vascular hyperpermeability and edema formation. The KDR tyrosine kinase may also be referred to as FLK-1 tyrosine kinase, NYK tyrosine kinase or VEGFR-2 tyrosine kinase. KDR tyrosine kinase is activated when vascular endothelial cell growth factor (VEGF) or another activating ligand (such as VEGF-C, VEGF-D, VEGF-E or HIV Tat protein) binds to a KDR tyrosine kinase receptor which lies on the surface of vascular endothelial cells. Following such KDR tyrosine kinase activation, hyperpermeability of the blood vessels occurs and fluid moves from the blood stream past the blood vessel walls into the interstitial spaces, thereby forming an area of edema. Diapedesis also often accompanies this response. Similarly, excessive vascular hyperpermeability can disrupt normal molecular exchange across the endothelium in critical tissues and organs (e.g., lung and kidney), thereby causing macromolecular extravasation and deposition. Following this acute response to KDR stimulation which is believed to facilitate the subsequent angiogenic process, prolonged KDR tyrosine kinase stimulation results in the proliferation and chemotaxis of vascular endothelial cells and formation of new vessels. By inhibiting KDR tyrosine kinase activity, either by blocking the production of the activating ligand, by blocking the activating ligand binding to the KDR tyrosine kinase receptor, by preventing receptor dimerization and transphosphorylation, by inhibiting the enzyme activity of the KDR tyrosine kinase (inhibiting the phosphorylation function of the enzyme) or by some other mechanism that interrupts its downstream signaling (D. Mukhopedhyay et al., Cancer Res. 58:1278-1284 (1998) and references therein), hyperpermeability, as well as associated extravasation, subsequent edema formation and matrix deposition, and angiogenic responses, may be inhibited and minimized.

Brief Summary Text - BSTX (170):

The method of the present invention is useful in the treatment of protein kinase-mediated conditions, such as any of the conditions described above. In one embodiment, the protein kinase-mediated condition is characterized by undesired angiogenesis, edema, or stromal deposition. For example, the condition can be one or more more ulcers, such as ulcers caused by bacterial or fungal infections, Mooren ulcers and ulcerative colitis. The condition can also be due to a microbial infection, such as Lyme disease, sepsis, septic shock or infections by Herpes simplex, Herpes Zoster, human immunodeficincy virus, protozoa, toxoplasmosis or parapoxvirus; an angiogenic disorders, such as von Hippel Lindau disease, polycystic kidney disease, pemphigoid, Paget's disease and psoriasis; a reproductive condition, such as endometriosis, ovarian hyperstimulation syndrome, preeclampsia or menometrorrhagia; a fibrotic and edemic condition, such as sarcoidosis, fibrosis, cirrhosis, thyroiditis, hyperviscosity syndrome systemic, Osler-Weber-Rendu disease, chronic occlusive

pulmonary disease, asthma, and edema following burns, trauma, radiation, stroke, hypoxia or ischemia; or an inflammatory/immunologic condition, such as systemic lupus, chronic inflammation, glomerulonephritis, synovitis, inflammatory bowel disease, Crohn's disease, rheumatoid arthritis, osteoarthritis, multiple sclerosis and graft rejection. Suitable protein kinase-mediated conditions also include sickle cell anaemia, osteoporosis, osteopetrosis, tumor-induced hypercalcemia and bone metastases. Additional protein kinase-mediated conditions which can be treated by the method of the present invention include ocular conditions such as ocular and macular edema, ocular neovascular disease, scleritis, radial keratotomy, uveitis, vitritis, myopia, optic pits, chronic retinal detachment, post-laser complications, conjunctivitis, Stargardt's disease and Eales disease, in addition to retinopathy and macular degeneration.

Brief Summary Text - BSTX (176):

In many pathological conditions (for example, solid primary tumors and metastases, Kaposi's sarcoma, rheumatoid arthritis, blindness due to inappropriate ocular neovascularization, psoriasis and atherosclerosis) disease progression is contingent upon persistent angiogenesis. Polypeptide growth factors often produced by the disease tissue or associated inflammatory cells, and their corresponding endothelial cell specific receptor tyrosine kinases (e.g., KDR/VEGFR-2, Flt-1/VEGFR-1, Tie-2/Tek and Tie) are essential for the stimulation of endothelial cell growth, migration, organization, differentiation and the establishment of the requisite new functional vasculature. As a result of the vascular permeability factor activity of VEGF in mediating vascular hyperpermeability, VEGF-stimulation of a VEGFR kinase is also believed to play an important role in the formation of tumor ascites, cerebral and pulmonary edema, pleural and pericardial effusions, delayed-type hypersensitivity reactions, tissue edema and organ dysfunction following trauma, burns, ischemia, diabetic complications, endometriosis, adult respiratory distress syndrome (ARDS), post-cardiopulmonary bypass-related hypotension and hyperpermeability, and ocular edema leading to glaucoma or blindness due to inappropriate neovascularization. In addition to VEGF, recently identified VEGF--C and VEGF-D, and virally-encoded VEGF-E or HIV-Tat protein can also cause a vascular hyperpermeability response through the stimulation of a VEGFR kinase. KDR/VEGFR-2 and/or Tie-2 are expressed also in a select population of hematopoietic stem cells. Certain members of this population are pluripotent in nature and can be stimulated with growth factors to differentiate into endothelial cells and participate in vasculogenetic angiogenic processes. For this reason these have been called Endothelial Progenitor Cells (EPCs) (J. Clin. Investig. 103: 1231-1236 (1999)). In some progenitors, Tie-2 may play a role in their recruitment, adhesion, regulation and differentiation (Blood, 4317-4326 (1997)). Certain agents according to formula I capable of blocking the kinase activity of endothelial cell specific kinases could therefore inhibit disease progression involving these situations.

Brief Summary Text - BSTX (178):

The compounds of formula I or a salt thereof or pharmaceutical compositions containing a therapeutically effective amount thereof may be used in the treatment of protein kinase-mediated conditions, such as benign and neoplastic proliferative diseases and disorders of the immune system, as described above. For example, such diseases include autoimmune diseases, such as rheumatoid arthritis, thyroiditis, type 1 diabetes, multiple sclerosis, sarcoidosis, inflammatory bowel disease, Crohn's disease, myasthenia gravis and systemic lupus erythematosus; psoriasis, organ transplant rejection (eg. kidney rejection, graft versus host disease), benign and neoplastic proliferative diseases, human cancers such as lung, breast, stomach, bladder, colon, pancreas, ovarian, prostate and rectal cancer and hematopoietic malignancies (leukemia and lymphoma), and diseases involving inappropriate vascularization

for example diabetic retinopathy, retinopathy of prematurity, choroidal neovascularization due to age-related macular degeneration, and infantile hemangiomas in human beings. In addition, such inhibitors may be useful in the treatment of disorders involving VEGF mediated edema, ascites, effusions, and exudates, including for example macular edema, cerebral edema, acute lung injury and adult respiratory distress syndrome (ARDS).

Brief Summary Text - BSTX (186):
The compounds of this invention can be administered to a human patient by themselves or in pharmaceutical compositions where they are mixed with suitable carriers or excipient(s) at doses to treat or ameliorate vascular hyperpermeability, edema and associated disorders. Mixtures of these compounds can also be administered to the patient as a simple mixture or in suitable formulated pharmaceutical compositions. A therapeutically effective dose further refers to that amount of the compound or compounds sufficient to result in the prevention or attenuation of inappropriate neovascularization, progression of hyperproliferative disorders, edema, VEGF-associated hyperpermeability and/or VEGF-related hypotension. Techniques for formulation and administration of the compounds of the instant application may be found in "Remington's Pharmaceutical Sciences," Mack Publishing Co., Easton, Pa., latest edition.

Brief Summary Text - BSTX (189):
Alternatively, one may administer the compound in a local rather than a systemic manner, for example, via injection of the compound directly into an edematous site, often in a depot or sustained release formulation.

Brief Summary Text - BSTX (230):
In the compositions of the present invention the active compound may, if desired, be associated with other compatible pharmacologically active ingredients. For example, the compounds of this invention can be administered in combination with one or more additional pharmaceutical agents that inhibit or prevent the production of VEGF or angiopoietins, attenuate intracellular responses to VEGF or angiopoietins, block intracellular signal transduction, inhibit vascular hyperpermeability, reduce inflammation, or inhibit or prevent the formation of edema or neovascularization. The compounds of the invention can be administered prior to, subsequent to or simultaneously with the additional pharmaceutical agent, whichever course of administration is appropriate. The additional pharmaceutical agents include but are not limited to anti-edemic steroids, NSAIDS, ras inhibitors, anti-TNF agents, anti-IL1 agents, antihistamines, PAF-antagonists, COX-1 inhibitors, COX-2 inhibitors, NO synthase inhibitors, Akt/PTB inhibitors, IGF-1R inhibitors, PKC inhibitors and PI3 kinase inhibitors. The compounds of the invention and the additional pharmaceutical agents act either additively or synergistically. Thus, the administration of such a combination of substances that inhibit angiogenesis, vascular hyperpermeability and/or inhibit the formation of edema can provide greater relief from the deletrious effects of a hyperproliferative disorder, angiogenesis, vascular hyperpermeability or edema than the administration of either substance alone. In the treatment of malignant disorders combinations with antiproliferative or cytotoxic chemotherapies or radiation are anticipated.

Brief Summary Text - BSTX (299):
In vivo Uterine Edema Model

Brief Summary Text - BSTX (300):
This assay measures the capacity of compounds to inhibit the acute increase in uterine weight in mice which occurs in the first few hours following estrogen stimulation. This early onset of uterine weight increase is known to

be due to edema caused by increased permeability of uterine vasculature. Cullinan-Bove and Koss (Endocrinology (1993), 133:829-837) demonstrated a close temporal relationship of estrogen-stimulated uterine edema with increased expression of VEGF mRNA in the uterus. These results have been confirmed by the use of neutralizing monoclonal antibody to VEGF which significantly reduced the acute increase in uterine weight following estrogen stimulation (WO 97/42187). Hence, this system can serve as a model for in vivo inhibition of VEGF signalling and the associated hyperpermeability and edema.

Brief Summary Text - BSTX (309):
Results demonstrate that certain compounds of the present invention inhibit the formation of edema when administered systemically by various routes.

Claims Text - CLTX (96):
64. A method of affecting angiogenesis or vascular permeability in a patient, comprising the step of administering to the patient a therapeutically effective amount of a compound of claim 1 or a physiologically acceptable salt thereof.

Claims Text - CLTX (101):
69. A method of treating a patient having a condition, comprising the step of administering to the patient a therapeutically effective amount of a compound of claim 1 wherein the condition is selected from the group consisting of one or more ulcers, an ulcer or ulcers caused by a bacterial or fungal infection, an ulcer or ulcers that are a symptom of ulcerative colitis, Lyme disease, sepsis, septic shock, infection by Herpes simplex, Herpes Zoster, human immunodeficiency virus, parapoxvirus, protozoa or toxopasmosis, von Hippel Lindau disease, pemphigoid, psoriasis, Paget's disease, polycycstic kidney disease, fibrosis, sarcoidosis, cirrhosis, thyroiditis, hyperviscosity syndrome, Osler-Weber-Rendu disease, chronic occlusive pulmonary disease, asthma, exudtaes, ascites, pleural effusions, pulmonary edema, cerebral edema or edema following bums, trauma, radiation, stroke, hypoxia, iscbemia, ovarian hyperstimulation syndrome, preeclampsia, menometrorrhagia, endometriosis, glomemlonephritis, synovitis, inflammatory bowel disease, rheumatoid arthritis, osteoarthritis, graft rejection, sickle cell anaemia, ocular or macular edema, ocular neovascular disease, scleritis, uveitis, vitritis, myopia, optic pits, chronic retinal detachment, post-laser treatment complications, conjunctivitis, Stargardt's disease, Eales disease, retinopathy, macular degeneration, atherosclerosis, restenosis, ischemia/reperfusion injury, vascular occlusion, venous malformation, carotid obstructive disease, fibrosarcoma, osteoma, melanoma, retinoblastoma, a rhabdomyosarcoma, glioblastoma, neuroblastoma, teratocarcinoma, hematopoietic malignancy, malignant ascites, Kaposi's sarcoma, Hodgkin's disease, lymphoma, myeloma, leukemia, Crow-Fukase (POEMS) syndrome, insulin-dependent diabetes mellitus glaucoma, diabetic retinopathy or microangiopathy.

Other Reference Publication - OREF (13):
Hanke, J.H., et al., "Discovery of a Novel, Potent, and Src Family-selective Tyrosine Kinase Inhibitor; Study of Lck- and FynT-dependent T Cell Activation," J Biol Chem., 271(2) :695-701, 1996.

Other Reference Publication - OREF (16):
Missbach, M., et al., "A Novel Inhibitor of the Tyrosine Kinase Src Suppresses Phosphorylation of Its Major Cellular Substrates and Reduces Bone Resorption In Vitro and in Rodent Models In Vivo," Bone, 24 (5) :437-449 (1999).

US-PAT-NO: 6713462

DOCUMENT-IDENTIFIER: US 6713462 B2

TITLE: Quinolinones and uses thereof

DATE-ISSUED: March 30, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Metcalf, III; Chester A.	Needham	MA	N/A	N/A
Shakespeare; William C.	Southborough	MA	N/A	N/A
Sawyer; Tomi K.	Southborough	MA	N/A	N/A
Wang; Yihan	Newton	MA	N/A	N/A
Bohacek; Regine	Boston	MA	N/A	N/A
Sundaramoorthi; Rajeswari	Watertown	MA	N/A	N/A

APPL-NO: 10/ 177500

DATE FILED: June 21, 2002

PARENT-CASE:

PRIORITY INFORMATION

The present application claims priority under 35 U.S.C. .sctn.119 to U.S. provisional application No. 60/299,936, filed Jun. 21, 2001, entitled "Novel Quinolinones and Uses Thereof", the entire contents of which are hereby incorporated by reference.

US-CL-CURRENT: 514/82, 514/312 , 546/153 , 546/155 , 546/157 , 546/158 , 546/23

ABSTRACT:

The invention relates to compounds of the general formula (and pharmaceutically acceptable derivatives thereof): ##STR1## in which R.sup.A, R.sup.B, R.sup.C, R.sup.D, R.sup.5, R.sup.7, R.sup.9, R.sup.9a, AK, p, q, r and X are as defined herein, and to their preparation and use.

75 Claims, 0 Drawing figures

Exemplary Claim Number: 1

---------- KWIC ---------

Brief Summary Text - BSTX (2):

The need to treat elusive and debilitating disorders such as cancer, osteoporosis and other diseases involving untoward bone resorption (e.g., Paget's Disease, primary and secondary hyperparathyroidism, humoral hypercalcemia of malignancy, various cancers where resorption is increased, and rheumatoid arthritis), and disorders involving increased vascular permeability, to name a few, has led to extensive research on the mechanisms involved in disease initiation and/or progression and on the identification of new drugs which might interfere with those mechanisms.

Brief Summary Text - BSTX (4):

Another approach to drug discovery for treating bone-related (and other) diseases involves the control of cellular signal transduction. See, for example, Missbach et al., "A Novel Inhibitor of the Tyrosine Kinase Src Suppresses Phosphorylation of Its Major Cellular Substrates and Reduces Bone Resorption In Vitro and in Rodent Models In Vivo." Bone 1999, 24, 437-449; Connolly et al., Bioorg. & Med. Chem. Lett. 1997, 7, 2415-2420; Trump-Kallmeyer et al., J. Med. Chem. 1998, 41, 1752-1763; Klutchko et al., J. Med. Chem. 1998, 41, 3276-3292; Legraverend et al., Bioorg. & Med. Chem. 1999, 7, 1281-1293; Chang et al., Chem. & Biol. 1999, 6, 361-375; Lev et al. Nature 1995, 376, 737-784; Palmer et al., J. Med. Chem. 1997, 40, 1519-1529.

Brief Summary Text - BSTX (7):

Protein kinases, specifically Src protein kinases, have been shown to play a crucial role in osteoclast function and thus in the resorption of bone and the progression of the osteoporosis. In addition, cellular signal transduction mediated by kinases like Src is believed to play a key role in other diseases, for example cancer and diseases involving increased vascular permeability. Though the exact mechanisms of signal transduction is still unclear, tyrosine kinases have been shown to be important contributing factors in cell proliferation, carcinogenesis and cell differentiation.

Brief Summary Text - BSTX (9):

Furthermore, certain kinases are believed to mediate signaling activity in response to a variety of growth factors, including VEGF, vascular endothelial growth factor, (see, Schlessinger, J. Cell 2000, 100, 293; Lowell et al. Genes Dev. 1996, 10, 1845), which is an angiogenic factor that promotes vascular permeability. The ability to control (and/or diminish) increased vascular permeability by suppression of a signalling pathway would be useful for the treatment of patients suffering from diseases and conditions related to increases in vascular permeability (e.g., edema, hemorrhage, cancer, vasular leaks, and the like). For a review of antiangiogenic agents (including those agents having antitumor activity), see Klohs et al., Curr. Opin. Biotechnol. 1999, 10, 544.

Brief Summary Text - BSTX (10):

Although some progress has been made in the treatment of certain debilitating diseases and disorders mentioned herein, there remains a need to develop new therapeutic agents which have an improved therapeutic index, which may be given to patients who cannot well tolerate or do not respond to existing therapies, and/or which may be used in conjunction with other therapies. Thus, new, selective inhibitors of osteoclast activity and promoters of osteoblast activity as well as therapeutic agents that can regulate a variety of other signal transduction pathways would be desirable. Such compounds may then be used to inhibit or promote complex biological processes in order to treat and/or prevent diseases associated with signalling (e.g., osteoporosis, cancer and edema, to name a few).

Brief Summary Text - BSTX (117):

This invention also provides a pharmaceutical preparation comprising at least one of the foregoing compounds or a pharmaceutically acceptable derivative thereof, as inhibitors of bone resorption by osteoclasts, as inhibitors of tumor growth and tumor metastatsis, for the treatment and prophylaxis of diseases or undesirable conditions which are mediated by a kinase inhibited by said compound, as inhibitors of vascular permeability and/or angiogenesis, and at least one pharmaceutically acceptable excipient or additive. Preferably the excipient of additive is pharmaceutically innocuous.

Brief Summary Text - BSTX (118):

The invention further provides a method for inhibiting bone resorption, inhibiting tumor growth and/or tumor metastasis, inhibiting vascular permeability and/or angiogenesis, or for the treatment and prevention of diseases or undesirable conditions which are mediated by a kinase inhibited by one of the foregoing compounds. The method involves administering a therapeutically effective amount of the compound or a pharmaceutically acceptable derivative thereof to a human or animal in need of it. Such administration constitutes a method for inhibiting bone resorption by osteoclasts, for inhibiting tumor growth and/or tumor metastasis or other proliferative disease, or for inhibiting vascular permeability and/or angiogenesis. Generally speaking, such administration comprises a method for the treatment and prophylaxis of diseases which are mediated by a kinase inhibited by one of the foregoing compounds or a pharmaceutically acceptable derivative thereof.

Brief Summary Text - BSTX (121):

This invention provides a new family of compounds with a range of biological properties. Compounds of this invention have biological activities relevant for the treatment of diseases including bone related disorders, disorders related to cellular proliferation (e.g., cancer) and disorders related to increased vascular permeability and/or angiogenesis. More generally, the compounds are useful in the regulation of signal transduction pathways. For example, certain compounds of the invention are useful for inhibiting tyrosine kinases, including without limitation receptor-type tyrosine kinases such as those of the HER (e.g. EGFR, HER2, HER3 and HER4), PDGF and FLK families (including, e.g., VEGF-R1 and VEGF-R2) as well as non-reecptor-type tyrosine kinases such as those of the Src and abl subfamilies, again as non-limiting examples.

Brief Summary Text - BSTX (126):

As described herein, compounds of the invention may be substituted with any number of substituents or functional groups, such as are illustrated in connection with particular classes, subclasses and species of the invention. In general, the term "substituted" and "substituent", whether preceded by the term "optionally" or not, refer to the replacement of hydrogen radicals in a given structure with the radical of a specified substituent. When more than one position in any given structure may be substituted with more than one substituent selected from a specified group, the substituent may be either the same or different at every position. As used herein, the term "substituted" encompasses all permissible substituents of organic compounds. Substituents are discussed in detail below and illustrated throughout this document. Combinations of substituents and variables envisioned by this invention are preferably those that result in the formation of stable compounds, useful in the treatment of various disorders as described herein, e.g. for bone related disorders, cancer or other disorders related to excessive cellular proliferation, disorders related to increases in vascular permeability, and/or more generally, disorders related to cell signalling. The term "stable", as used herein, refers to compounds that possess stability sufficient to allow their production, detection and preferably their recovery, purification and use for one or more of the purposes disclosed herein.

Brief Summary Text - BSTX (148):

The inclusion of a phosphorus-containing moiety in the design of the compounds of this invention can impart interesting functional characteristics to the compounds. For instance, depending in some cases on the choice of phosphorus-containing moiety and/or its location in the compound, characteristics of the compounds such as in vitro or in vivo potency, ClogP, aqueous solubility, ability to penetrate cells, and ability to target bone tissue may be desirably affected. As discussed above the novel compounds of

this invention have biological properties which make them of interest for the treatment of bone disorders, disorders related to cellular proliferation (e.g., cancer), and disorders resulting from increased vascular permeability and/or angiogenesis.

Brief Summary Text - BSTX (150):
In certain embodiments, these compositions optionally further comprise, or are administered conjointly with, one or more additional therapeutic agents. For example, the additional therapeutic agent may be an anticancer agent, an agent for the treatment of a bone disorder, or an agent for the treatment of disorders related to increased vascular permeability and/or angiogenesis, as discussed in more detail herein.

Brief Summary Text - BSTX (157):
Thus, administering to a subject in need thereof a therapeutically effective amount of a compound of the invention, or a composition containing such compound or a pharmaceutically acceptable derivative thereof, provides a method for the treatment of those disorders. A "therapeutically effective amount" is an amount effective for detectably ameliorating the disorder, e.g., an amount effective for detectably killing or inhibiting the growth of tumor cells; for inhibiting osteoclast activity, slowing bone resorption, increasing bone growth or reducing serum calcium levels; or for inhibiting antiangiogenesis or edema or a manifestation thereof.

Brief Summary Text - BSTX (174):
As discussed above, in another aspect, the compounds of this invention are useful in the selective treatment or prevention of bone disorders, and may effect treatment via inhibition of osteoclast activity, promotion of osteoblast activity, or promotion or inhibition of other cellular events necessary for healthy bone metabolism. In certain preferred embodiments, these compounds are useful for the treatment or prevention of diseases and conditions associated with bone metabolic disorders such as osteoclast overactivity. In still other embodiments, the compounds of this invention are targeted Src kinase inhibitors and thus inhibit bone resorption by osteoclasts.

Brief Summary Text - BSTX (176):
In a further aspect, the present invention provides an inhibitor of mammalian osteoclasts, for example any one of the compounds of this invention or a pharmaceutical composition thereof. In still another aspect, the present invention provides compounds or pharmaceutical compositions that are selective Src kinase inhibitors. In particular, the method of present invention comprises providing any one of the compounds of this invention or a pharmaceutically composition thereof, for use in the treatment of and/or prophylaxis of osteoporosis and related osteopenic diseases.

Brief Summary Text - BSTX (178):
In yet another embodiment, in addition to the treatment or prevention of osteoporosis or cancer, the present invention can be utilized to inhibit increases in vascular permeability. For example, certain compounds are tested for the ability to inhibit the tyrosine kinase activity associated with the VEGF receptors such as Flt and/or KDR and for their ability to inhibit angiogenesis and/or increased vascular permeability. Additionally, these compounds can be tested for the ability to inhibit the tyrosine kinase activity associated with Src and for their ability to inhibit angiogenesis and/or increased vascular permeability. These properties may be assessed, for example, using one or more of the procedures set out below. Thus according to this aspect of the invention there is provided a method for reducing vascular permability in a subject comprising administering a compound of Formula I, as described herein and as described by the various classes and subclasses.

Brief Summary Text - BSTX (179):

It will be appreciated that, similarly to the anticancer treatment and treatment for osteoporosis, as also described herein, the antiangiogenic and/or vascular permeability reducing treatment defined herein may be applied as a sole therapy or may involve, in addition to a compound of the invention, one or more other substances and/or treatments. Such conjoint treatment may be achieved by way of the simultaneous, sequential or separate administration of the individual components of the treatment. In the field of medical oncology it is normal practice to use a combination of different forms of treatment to treat each patient with cancer. In medical oncology the other component(s) of such conjoint treatment in addition to the antiangiogenic end/or vascular permeability reducing treatment defined herein may be: surgery, radiotherapy or chemotherapy. Such chemotherapy may cover three main categories of therapeutic agent: (1) other antiangiogenic agents that work by different mechanisms from those defined hereinbefore (for example linomide, angiostatin, razoxin, thalidomide); (ii) cytostatic agents such as antioestrogens (for example tamoxifen, toremifene, raloxifene, droloxifene, iodoxyfene), progestogens (for example megestrol acetate), aromatase inhibitors (for example anastrozole, letrazole, vorazole, exemestane), antiprogestogens, antiandrogens (for example flutamide, nilutamide, bicalutamide, cyproterone acetate), LHRH agonists and antagonists (for example goserelin acetate, luprolide), inhibitors of testosterone 5.alpha.-dihydroreductase (for example finasteride), anti-invasion agents (for example metalloproteinase inhibitors like marimastat and inhibitors of urokinase plasminogen activator receptor function) and inhibitors of growth factor function, (such growth factors include for example EGF, FGFs, platelet derived growth factor and hepatocyte growth factor such inhibitors include growth factor antibodies, growth factor receptor antibodies, tyrosine kinase inhibitors and serine/threonine kinase inhibitors); and (iii) antiproliferative/antineoplastic drugs and combinations thereof, as used in medical oncology, such as antimetabolites (for example antifolates like methotrexate, fluoropyrimidines like 5-fluorouracil, purine and adenosine analogues, cytosine arabinoside); antitumour antibiotics (for example anthracyclines like doxorubicin, daunomycin, epirubicin and idarubicin, mitomycin-C, dactinomycin, mithramycin); platinum derivatives (for example cisplatin, carboplatin); alkylating agents (for example nitrogen mustard, melphalan, chlorambucil, busulphan, cyclophosphamide, ifosfamide, nitrosoureas, thiotepa); antimitotic agents (for example vinca alkaloids like vincristine and taxoids like taxol, taxotere); topoisomerase inhibitors (for example epipodophyllotoxins like etoposide and teniposide, amsacrine, topotecan).

Brief Summary Text - BSTX (180):

As stated above, in another embodiment of the invention, the compounds defined in the present invention are of interest for their antiangiogenic and/or vascular permeability reducing effects. Such compounds of the invention are expected to be useful in a wide range of disease states including cancer, diabetes, psoriasis, rheumatoid arthritis, Kaposi's sarcoma, haemangioma, acute and chronic nephropathies, atheroma, arterial restenosis, autoimmune diseases, acute inflammation and ocular diseases with retinal vessel proliferation. In particular, such compounds of the invention are expected to slow advantageously the growth of primary and recurrent solid tumours of, for example, the colon, breast, prostate, lungs and skin. More particularly such compounds of the invention are expected to inhibit the growth of those primary and recurrent solid tumours which are associated with VEGF, especially those tumours which are significantly dependent on VEGF for their growth and spread, including for example, certain tumours of the colon, breast, prostate, lung, vulva and skin.

Detailed Description Text - DETX (147):

Compounds of the present invention may be evaluated in a variety of assays

to determine or characterize their biological activities. For example, the compounds of the invention can be tested for their ability to inhibit Src kinase or other protein kinases, to bind to bone, to inhibit bone resorption or to otherwise improve the relative dynamics of bone homeostasis. The compounds can also be evaluated for their cytotoxic and growth inhibitory effects on tumor cells of interest. Furthermore, the compounds can be evaluated for their ability to act as vitronectin receptor antagonists and as inhibitors of cell adhesion.

Detailed Description Text - DETX (156):

A murine hypercalcemia model for determining the efficacy of Src kinase inhibitors was developed. This model exploits the intrinsic effects of PTH (1-34) to stimulate the resorptive activity of osteoclasts in vivo. Briefly, compounds are each injected into mice subcutaneously, once or twice per day for five consecutive days. On the third day of test compound treatments, PTH administration begins. PTH (20 .mu.g/kg) is given four times per day, subcutaneously, until the end of the study. Control animals receive PTH but do not receive test compounds. Blood samples are collected from the animals to obtain baseline (pre-PTH treatment), 48 hour and 72 hour (after initiation of PTH treatment) serum samples. The serum samples are analyzed for calcium concentration using the quantitative calorimetric assay reagent Arsenazo III (Sigma). Calcium serum levels for treated groups are compared to calcium serum levels of control groups and a percentage of inhibition of hypercalcemia is calculated for each time point. When a compound is effective in inhibiting the activity of osteoclasts, observed serum calcium concentrations are lower than those in animals that receive only PTH in the absence of test compound.

Detailed Description Text - DETX (158):

In addition to their ability to inhibit bone resorption, the compounds of the present invention are also able to inhibit protein kinase activity. For example, inventive compounds can be assessed for their ability to inhibit the activity of receptor and non-receptor tyrosine protein kinases. For example, the present invention presents a general method for determining the ability inhibit the activity of non-receptor tyrosine protein kinases (e.g., members of the src family, abl kinase, and ZAP70 kinase) and receptor tyrosine protein kinases (e.g., EGF family (c-erbB2, c-erbB3, and c-erbB4), the PDGF family (e.g., PDGF receptor, CSF-1, Kit, VEGF and FGF). Thus, the inventive compounds can be used in the immunomodulation and in the treatment of diseases of the immune system, for example in the case of inflammations or organ transplants. They are also suitable for the treatment of hyperproliferative diseases, including, but not limited to psoriasis, tumors, carcinomas and leukemias, and in fibrosis and restenosis. Additionally, compounds can be utilized for the treatment of diseases of the central or the peripheral nervous system where signal transmission by at least one tyrosine protein kinase is involved. Furthermore, Src and certain other kinases are believed to mediate signaling activity in response to a variety of growth factors, including VEGF, vascular endothelial growth factor, which is is an angiogenic factor that promotes vascular permeability, and thus certain inhibitors are usefulas antiangiogenic agents. In addition to the kinase assays described in this section, certain other kinase assays are described in the context of anti-angiogenic agents below, for example.

Detailed Description Text - DETX (173):

a) Src Kinase Inhibition Assay:

Detailed Description Text - DETX (174):

Compounds are tested for their ability to inhibit Src kinase using the scintillation proximity assay (SPA) technology as developed by Amersham. Reagents include: Streptavidin SPA beads from Amersham,

2-[N-morpholino]ethanesulfonic acid from Sigma, ATP from Boerhinger Mannheim, [.sup.33 P]ATP: from NEN (NEG 602H), the substrate--biotinylated peptide substrate 1 (PKS1) (cdc2 peptide) from Pierce which is prepared at 12.5 .mu.M (5.times.solution) in kinase buffer, and the enzyme, human recombinant c-Src at 135 .mu.g/ml (stock solution) which is diluted 1/40 in kinase buffer (3.38 .mu.g/ml) before use. Buffers include: (a) Kinase buffer which contains MES 30 mM pH 6.8, MgCl.sub.2 10 mM, Orthovanadate 0.25 mM, PMSF 0.1 mM, and DTT 1 mM; (b) ATP buffer which contains ATP 5 mM in MgCl.sub.2 50 mM buffer (stock solution). Note that before each use dilute in MES to 100 .mu.M (5.times.solution) add 100 .mu.Ci/mL [.sup.33 P]ATP; and (c) PBS Stop buffer which contains ATP 0.1 mM, EDTA 40 mM, Triton 0.1%. Streptavidin beads are suspended at 3.3 mg/ml in stop buffer and mixed by shaking. The Kinase reaction proceeds by stepwise addition to wells on the 96 well-plate of the following: (a) 10 .mu.L kinase buffer+10% DMSO or compound to be tested at different concentration in MES+10% DMSO, (b) 10 .mu.L kinase buffer, (c) 10 .mu.L substrate 12.5 .mu.M, (d) 10 .mu.L enzyme 3.38 .mu.g/ml, and (e) 10 .mu.L ATP 100 .mu.M containing 0.2 .mu.Ci [.sup.33 P]ATP. Incubation for 2 hours at 30 degrees C. is followed by addition of 150 .mu.L Stop buffer containing 500 .mu.g streptavidin beads. Incubation proceeds for 30 min at room temperature, followed by centrifugation for 5 min at 2000 rpm, and reading on a Wallac Microbeta Scintillation counter.

Detailed Description Text - DETX (192):
F. Vascular Permeability:

Detailed Description Text - DETX (193):
As mentioned above, certian kinases are believed to mediate signaling activity in response to a variety of growth factors, including VEGF, vascular endothelial growth factor, which is an angiogenic factor that promotes vascular permeability. For example, certain compounds are tested for the ability to inhibit the tyrosine kinase activity associated with the VEGF receptors such as Flt and/or KDR and for their ability to inhibit angiogenesis and/or increased vascular permeability. Additionally, these compounds can be tested for the ability to inhibit the tyrosine kinase activity associated with Src and for their ability to inhibit angiogenesis and/or increased vascular permeability. These properties may be assessed, for example, using one or more of the procedures set out below:

Detailed Description Text - DETX (204):
This test measures the capacity of compounds to reduce the acute increase in uterine weight in rats which occurs in the first 4-6 hours following oestrogen stimulation. This early increase in uterine weight has long been known to be due to oedema caused by increased permeability of the uterine vasculature and recently Cullinan-Bove and Koos (Endocrinology, 1993, 133:829-837) demonstrated a close temporal relationship with increased expression of VEGF mRNA in the uterus. It has been found that prior treatment of the rats with a neutralising monoclonal antibody to VEGF significantly reduces the acute increase in uterine weight, confirming that the increase in weight is substantially mediated by VEGF.

Claims Text - CLTX (69):
69. The composition of claim 68, wherein the composition further comprises an additional therapeutic agent and the therapeutic agent is an anticancer agent, an antiproliferative agent, an approved agent for the treatment of osteoporosis, or an approved agent for the treatment of disorders related to increased vascular permeability.

Other Reference Publication - OREF (2):
Boschelli, D.H. et al. Synthesis and Src Kinase Inhibitory Activity of a

Series of 4-phenylamino-3-quinolinecarbonitriles. J. Med. Chem. Jan. 2001, vol. 44, pp. 822-833, see entire document.

Other Reference Publication - OREF (3):
Database CA on STN, Chemical Abstracts, (Columbus, OH USA), No. 134:100834, Wang Y.D. "Inhibitors of Src tyrosine kinase: the preparation and structure-activity relationship of 4-anilino-3-cyanoquinolines and 4-anilinoquinazolines," abstract, Bioorganic & Medicinal Chemistry Letters, 2000, vol. 10, No. 21, pp. 2477-2480.

US-PAT-NO: 6710067

DOCUMENT-IDENTIFIER: US 6710067 B2

TITLE: Mannich base prodrugs of 3-(pyrrol-2-ylmethylidene)-2-indolinone derivatives

DATE-ISSUED: March 23, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Moon; Malcolm Wilson	Kalamazoo	MI	N/A	N/A
Morozowich; Walter	Kalamazoo	MI	N/A	N/A
Gao; Ping	Portage	MI	N/A	N/A
Tang; Peng Cho	Moraga	CA	N/A	N/A

APPL-NO: 09/ 863804

DATE FILED: May 24, 2001

PARENT-CASE:

CROSS-REFERENCE

This application claims priority under 35 U.S.C. 119(e) to U.S. Provisional applications Ser. No. 60/207,000 filed on May 24, 2000, and Ser. No. 60/225,045, filed on Aug. 11, 2000, the disclosures of which are incorporated herein by reference in their entirety.

US-CL-CURRENT: 514/414, 548/468

ABSTRACT:

The present invention is directed to Mannich base prodrugs of certain 3-(pyrrol-2-ylmethylidene)-2-indolinone derivatives that modulate the activity of protein kinases ("PKs"). Pharmaceutical compositions comprising these compounds, methods of treating diseases related to abnormal PK activity utilizing pharmaceutical compositions comprising these compounds and methods of preparing them are also disclosed.

9 Claims, 0 Drawing figures

Exemplary Claim Number: 1

---------- KWIC ---------

Detailed Description Text - DETX (503):
This assay is used to screen for inhibitors of the tyrosine kinase Src.

Detailed Description Text - DETX (550):
Vascular Permeability Assay

Detailed Description Text - DETX (551):
Increased vascular permeability in tumor-dependent angiogenesis is due to a loosening of gap junctions in response to vascular endothelial growth factor (VEGF). The Miles assay for vascular permeability (Miles and Miles, J.

Physiol. 118: 228-257 (1952)) has been adapted to athymic mice in order to evaluate the ability of the compounds of the present invention to inhibit VEGF-induced vascular permeability in vivo.

US-PAT-NO: 6706699

DOCUMENT-IDENTIFIER: US 6706699 B2

TITLE: Quinolines and uses thereof

DATE-ISSUED: March 16, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Wang; Yihan	Newton	MA	N/A	N/A
Metcalf, III; Chester A.	Needham	MA	N/A	N/A
Shakespeare; William C.	Southborough	MA	N/A	N/A
Sawyer; Tomi K.	Southborough	MA	N/A	N/A
Bohacek; Regine	Boston	MA	N/A	N/A
Sundaramoorthi; Rajeswari	Watertown	MA	N/A	N/A

APPL-NO: 10/ 177990

DATE FILED: June 21, 2002

PARENT-CASE:

PRIORITY INFORMATION

The present application claims priority under 35 U.S.C. .sctn.119 to U.S. provisional application No. 60/299,918, filed Jun. 21, 2001, entitled "Novel Quinolines and Uses Thereof", the entire contents of which are hereby incorporated by reference.

US-CL-CURRENT: 514/82, 514/312 , 514/313 , 546/153 , 546/159 , 546/162 , 546/23

ABSTRACT:

This invention relates to compounds of the general formula: ##STR1##

in which R.sup.A, R.sup.B, R.sup.C and R.sup.D are as defined herein, and to their preparation and use.

44 Claims, 0 Drawing figures

Exemplary Claim Number: 1

---------- KWIC ---------

Brief Summary Text - BSTX (2):

The need to treat elusive and debilitating disorders such as cancer, osteoporosis and other diseases involving untoward bone resorption (e.g., Paget's Disease, primary and secondary hyperparathyroidism, humoral hypercalcemia of malignancy, various cancers where resorption is increased, and rheumatoid arthritis), and disorders involving increased vascular permeability, to name a few, has led to extensive research on the mechanisms involved in disease initiation and/or progression and on the identification of new drugs which might interfere with those mechanisms.

Brief Summary Text - BSTX (4):

Another approach to drug discovery for treating bone-related (and other) diseases involves the control of cellular signal transduction. See, for example, Missbach et al., "A Novel Inhibitor of the Tyrosine Kinase Src Suppresses Phosphorylation of Its Major Cellular Substrates and Reduces Bone Resorption In Vitro and in Rodent Models In Vivo." Bone 1999, 24, 437-449; Connolly et al., Bioorg. & Med. Chem. Lett. 1997, 7, 2415-2420; Trump-Kallmeyer et al., J. Med. Chem. 1998, 41, 1752-1763; Klutchko et al., J. Med. Chem. 1998, 41, 3276-3292; Legraverend et al., Bioorg. & Med. Chem. 1999, 7, 1281-1293; Chang et al., Chem. & Biol. 1999, 6, 361-375; Lev et al. Nature 1995, 376, 737-784; Palmer et al., J. Med. Chem. 1997, 40, 1519-1529.

Brief Summary Text - BSTX (7):

Protein kinases, specifically Src protein kinases, have been shown to play a crucial role in osteoclast function and thus in the resorption of bone and the progression of the osteoporosis. In addition, cellular signal transduction mediated by kinases like Src is believed to play a key role in other diseases, for example cancer and diseases involving increased vascular permeability. Though the exact mechanisms of signal transduction is still unclear, tyrosine kinases have been shown to be important contributing factors in cell proliferation, carcinogenesis and cell differentiation.

Brief Summary Text - BSTX (9):

Furthermore, certain kinases are believed to mediate signaling activity in response to a variety of growth factors, including VEGF, vascular endothelial growth factor, (see, Schlessinger, J. Cell 2000, 100, 293; Lowell et al. Genes Dev. 1996, 10, 1845), which is an angiogenic factor that promotes vascular permeability. The ability to control (and/or diminish) increased vascular permeability by suppression of a signalling pathway would be useful for the treatment of patients suffering from diseases and conditions related to increases in vascular permeability (e.g., edema, hemorrhage, cancer, vasular leaks, and the like). For a review of antiangiogenic agents (including those agents having antitumor activity), see Klohs et al., Curr. Opin. Biotechnol. 1999, 10, 544.

Brief Summary Text - BSTX (10):

Although some progress has been made in the treatment of certain debilitating diseases and disorders mentioned herein, there remains a need to develop new therapeutic agents which have an improved therapeutic index, which may be given to patients who cannot well tolerate or do not respond to existing therapies, and/or which may be used in conjunction with other therapies. Thus, new, selective inhibitors of osteoclast activity and promoters of osteoblast activity as well as therapeutic agents that can regulate a variety of other signal transduction pathways would be desirable. Such compounds may then be used to inhibit or promote complex biological processes in order to treat and/or prevent diseases associated with signalling (e.g., osteoporosis, cancer and edema, to name a few).

Detailed Description Text - DETX (327):

This invention also provides a pharmaceutical preparation comprising at least one of the foregoing compounds or a pharmaceutically acceptable derivative thereof, as inhibitors of bone resorption by osteoclasts, as inhibitors of tumor growth and tumor metastatsis, for the treatment and prophylaxis of diseases or undesirable conditions which are mediated by a kinase inhibited by said compound, as inhibitors of vascular permeability and/or angiogenesis, and at least one pharmaceutically acceptable excipient or additive. Preferably the excipient of additive is pharmaceutically innocuous.

Detailed Description Text - DETX (328):

The invention further provides a method for inhibiting bone resorption, inhibiting tumor growth and/or tumor metastasis, inhibiting vascular permeability and/or angiogenesis, or for the treatment and prevention of diseases or undesirable conditions which are mediated by a kinase inhibited by one of the foregoing compounds. The method involves administering a therapeutically effective amount of the compound or a pharmaceutically acceptable derivative thereof to a human or animal in need of it. Such administration constitutes a method for inhibiting bone resorption by osteoclasts, for inhibiting tumor growth and/or tumor metastasis or other proliferative disease, or for inhibiting vascular permeability and/or angiogenesis. Generally speaking, such administration comprises a method for the treatment and prophylaxis of diseases which are mediated by a kinase inhibited by one of the foregoing compounds or a pharamceutically acceptable derivative thereof.

Detailed Description Text - DETX (331):

This invention provides a new family of compounds with a range of biological properties. Compounds of this invention have biological activities relevant for the treatment of diseases including bone related disorders, disorders related to cellular proliferation (e.g., cancer) and disorders related to increased vascular permeability and/or angiogenesis. More generally, the compounds are useful in the regulation of signal transduction pathways. For example, certain compounds of the invention are useful for inhibiting tyrosine kinases, including without limitation receptor-type tyrosine kinases such as those of the HER (e.g. EGFR, HER2, HER3 and HER4), PDGF and FLK families (including, e.g., VEGF-R1 and VEGF-R2) as well as non-reecptor-type tyrosine kinases such as those of the Src and abl subfamilies, again as non-limiting examples.

Detailed Description Text - DETX (336):

As described herein, compounds of the invention may be substituted with any number of substituents or functional groups, such as are illustrated in connection with particular classes, subclasses and species of the invention. In general, the term "substituted" and "substituent", whether preceded by the term "optionally" or not, refer to the replacement of hydrogen radicals in a given structure with the radical of a specified substituent. When more than one position in any given structure may be substituted with more than one substituent selected from a specified group, the substituent may be either the same or different at every position. As used herein, the term "substituted" encompasses all permissible substituents of organic compounds. Substituents are discussed in detail below and illustrated throughout this document. Combinations of substituents and variables envisioned by this invention are preferably those that result in the formation of stable compounds, useful in the treatment of various disorders as described herein, e.g. for bone related disorders, cancer or other disorders related to excessive cellular proliferation, disorders related to increases in vascular permeability, and/or more generally, disorders related to cell signalling. The term "stable", as used herein, refers to compounds that possess stability sufficient to allow their production, detection and preferably their recovery, purification and use for one or more of the purposes disclosed herein.

Detailed Description Text - DETX (360):

The inclusion of a phosphorus-containing moiety in the design of the compounds of this invention can impart interesting functional characteristics to the compounds. For instance, depending in some cases on the choice of phosphorus-containing moiety and/or its location in the compound, characteristics of the compounds such as in vitro or in vivo potency, ClogP, aqueous solubility, ability to penetrate cells, and ability to target bone tissue may be desirably affected. As discussed above the novel compounds of

this invention have biological properties which make them of interest for the treatment of bone disorders, disorders related to cellular proliferation (e.g., cancer), and disorders resulting from increased vascular permeability and/or angiogenesis.

Detailed Description Text - DETX (362):
In certain embodiments, these compositions optionally further comprise, or are administered conjointly with, one or more additional therapeutic agents. For example, the additional therapeutic agent may be an anticancer agent, an agent for the treatment of a bone disorder, or an agent for the treatment of disorders related to increased vascular permeability and/or angiogenesis, as discussed in more detail herein.

Detailed Description Text - DETX (369):
Thus, administering to a subject in need thereof a therapeutically effective amount of a compound of the invention, or a composition containing such compound or a pharmaceutically acceptable derivative thereof, provides a method for the treatment of those disorders. A "therapeutically effective amount" is an amount effective for detectably ameliorating the disorder, e.g., an amount effective for detectably killing or inhibiting the growth of tumor cells; for inhibiting osteoclast activity, slowing bone resorption, increasing bone growth or reducing serum calcium levels; or for inhibiting antiangiogenesis or edema or a manifestation thereof.

Detailed Description Text - DETX (386):
As discussed above, in another aspect, the compounds of this invention are useful in the selective treatment or prevention of bone disorders, and may effect treatment via inhibition of osteoclast activity, promotion of osteoblast activity, or promotion or inhibition of other cellular events necessary for healthy bone metabolism. In certain preferred embodiments, these compounds are useful for the treatment or prevention of diseases and conditions associated with bone metabolic disorders such as osteoclast overactivity. In still other embodiments, the compounds of this invention are targeted Src kinase inhibitors and thus inhibit bone resorption by osteoclasts.

Detailed Description Text - DETX (388):
In a further aspect, the present invention provides an inhibitor of mammalian osteoclasts, for example any one of the compounds of this invention or a pharmaceutical composition thereof. In still another aspect, the present invention provides compounds or pharmaceutical compositions that are selective Src kinase inhibitors. In particular, the method of present invention comprises providing any one of the compounds of this invention or a pharmaceutically composition thereof, for use in the treatment of and/or prophylaxis of osteoporosis and related osteopenic diseases.

Detailed Description Text - DETX (390):
In yet another embodiment, in addition to the treatment or prevention of osteoporosis or cancer, the present invention can be utilized to inhibit increases in vascular permeability. For example, certain compounds are tested for the ability to inhibit the tyrosine kinase activity associated with the VEGF receptors such as Flt and/or KDR and for their ability to inhibit angiogenesis and/or increased vascular permeability. Additionally, these compounds can be tested for the ability to inhibit the tyrosine kinase activity associated with Src and for their ability to inhibit angiogenesis and/or increased vascular permeability. These properties may be assessed, for example, using one or more of the procedures set out below. Thus according to this aspect of the invention there is provided a method for reducing vascular permability in a subject comprising administering a compound of Formula I, as described herein and as described by the various classes and subclasses.

Detailed Description Text - DETX (391):
It will be appreciated that, similarly to the anticancer treatment and treatment for osteoporosis, as also described herein, the antiangiogenic and/or vascular permeability reducing treatment defined herein may be applied as a sole therapy or may involve, in addition to a compound of the invention, one or more other substances and/or treatments. Such conjoint treatment may be achieved by way of the simultaneous, sequential or separate administration of the individual components of the treatment. In the field of medical oncology it is normal practice to use a combination of different forms of treatment to treat each patient with cancer. In medical oncology the other component(s) of such conjoint treatment in addition to the antiangiogenic and/or vascular permeability reducing treatment defined herein may be: surgery, radiotherapy or chemotherapy. Such chemotherapy may cover three main categories of therapeutic agent:

Detailed Description Text - DETX (393):
As stated above, in another embodiment of the invention, the compounds defined in the present invention are of interest for their antiangiogenic and/or vascular permeability reducing effects. Such compounds of the invention are expected to be useful in a wide range of disease states including cancer, diabetes, psoriasis, rheumatoid arthritis, Kaposi's sarcoma, haemangioma, acute and chronic nephropathies, atheroma, arterial restenosis, autoimmune diseases, acute inflammation and ocular diseases with retinal vessel proliferation. In particular, such compounds of the invention are expected to slow advantageously the growth of primary and recurrent solid tumours of, for example, the colon, breast, prostate, lungs and skin. More particularly such compounds of the invention are expected to inhibit the growth of those primary and recurrent solid tumours which are associated with VEGF, especially those tumours which are significantly dependent on VEGF for their growth and spread, including for example, certain tumours of the colon, breast, prostate, lung, vulva and skin.

Detailed Description Text - DETX (546):
Compounds of this invention may be evaluated in a variety of assays to determine or characterize their biological activities. For example, the compounds of the invention can be tested for their ability to inhibit protein kinases (e.g., Src, EGF or VEGF). In certain embodiments, compounds can be tested for their ability to bind to bone, to inhibit bone resorption or to otherwise improve the relative dynamics of bone homeostasis. The compounds can also be evaluated for their cytotoxic and growth inhibitory effects on tumor cells of interest. Furthermore, the compounds can be evaluated for their ability to act as vitronectin receptor antagonists and as inhibitors of cell adhesion.

Detailed Description Text - DETX (555):
A murine hypercalcemia model for determining the efficacy of Src kinase inhibitors was developed. This model exploits the intrinsic effects of PTH (1-34) to stimulate the resorptive activity of osteoclasts in vivo. Briefly, compounds are each injected into mice subcutaneously, once or twice per day for five consecutive days. On the third day of test compound treatments, PTH administration begins. PTH (20 .mu.g/kg) is given four times per day, subcutaneously, until the end of the study. Control animals receive PTH but do not receive test compounds. Blood samples are collected from the animals to obtain baseline (pre-PTH treatment), 48 hour and 72 hour (after initiation of PTH treatment) serum samples. The serum samples are analyzed for calcium concentration using the quantitative colorimetric assay reagent Arsenazo III (Sigma). Calcium serum levels for treated groups are compared to calcium serum levels of control groups and a percentage of inhibition of hypercalcemia is calculated for each time point. When a compound is effective in inhibiting the

activity of osteoclasts, observed serum calcium concentrations are lower than those in animals that receive only PTH in the absence of test compound.

Detailed Description Text - DETX (557):

In addition to their ability to inhibit bone resorption, the compounds of this invention are also able to inhibit protein kinase activity. For example, inventive compounds can be assessed for their ability to inhibit the activity of receptor and non-receptor tyrosine protein kinases. For example, the present invention presents a general method for determining the ability inhibit the activity of non-receptor tyrosine protein kinases (e.g., members of the src family, abl kinase, and ZAP70 kinase) and receptor tyrosine protein kinases (e.g., EGF family (c-erbB2, c-erbB3, and c-erbB4), the PDGF family (e.g., PDGF receptor, CSF-1, Kit, VEGF and FGF). Thus, the inventive compounds can be used in the immunomodulation and in the treatment of diseases of the immune system, for example in the case of inflammations or organ transplants. They are also suitable for the treatment of hyperproliferative diseases, including, but not limited to psoriasis, tumors, carcinomas and leukemias, and in fibrosis and restenosis. Additionally, compounds can be utilized for the treatment of diseases of the central or the peripheral nervous system where signal transmission by at least one tyrosine protein kinase is involved. Furthermore, Src and certain other kinases are believed to mediate signaling activity in response to a variety of growth factors, including VEGF, vascular endothelial growth factor, which is is an angiogenic factor that promotes vascular permeability, and thus certain inhibitors are usefulas antiangiogenic agents. In addition to the kinase assays described in this section, certain other kinase assays are described in the context of anti-angiogenic agents below, for example.

Detailed Description Text - DETX (572):

a) Src Kinase Inhibition Assay:

Detailed Description Text - DETX (573):

Compounds are tested for their ability to inhibit Src kinase using the scintillation proximity assay (SPA) technology as developed by Amersham. Reagents include: Streptavidin SPA beads from Amersham, 2-[N-morpholino]ethanesulfonic acid from Sigma, ATP from Boerhinger Mannheim, [.sup.33 P]ATP: from NEN (NEG 602H), the substrate--biotinylated peptide substrate 1 (PKS1) (cdc2 peptide) from Pierce which is prepared at 12.5 .mu.M (5.times.solution) in kinase buffer, and the enzyme, human recombinant c-Src at 135 .mu.g/ml (stock solution) which is diluted 1/40 in kinase buffer (3.38 .mu.g/ml) before use. Buffers include: (a) Kinase buffer which contains MES 30 mM pH 6.8, MgCl.sub.2 10 mM, Orthovanadate 0.25 mM, PMSF 0.1 mM, and DTT 1 mM; (b) ATP buffer which contains ATP 5 mM in MgCl.sub.2 50 mM buffer (stock solution). Note that before each use dilute in MES to 100 .mu.M (5.times.solution) add 100 .mu.Ci/mL [.sup.33 P]ATP; and (c) PBS Stop buffer which contains ATP 0.1 mM, EDTA 40 mM, Triton 0.1%. Streptavidin beads are suspended at 3.3 mg/ml in stop buffer and mixed by shaking. The Kinase reaction proceeds by stepwise addition to wells on the 96 well-plate of the following: (a) 10 .mu.L kinase buffer+10% DMSO or compound to be tested at different concentration in MES+10% DMSO, (b) 10 .mu.L kinase buffer, (c) 10 .mu.L substrate 12.5 .mu.M, (d) 10 .mu.L enzyme 3.38 .mu.g/ml, and (e) 10 .mu.L ATP 100 .mu.M containing 0.2 .mu.Ci [.sup.33 P]ATP. Incubation for 2 hours at 30 degrees C. is followed by addition of 150 .mu.L Stop buffer containing 500 .mu.g streptavidin beads. Incubation proceeds for 30 min at room temperature, followed by centrifugation for 5 min at 2000 rpm, and reading on a Wallac Microbeta Scintillation counter.

Detailed Description Text - DETX (595):

e) Vascular Permeability:

Detailed Description Text - DETX (596):

As mentioned above, certian kinases are believed to mediate signaling activity in response to a variety of growth factors, including VEGF, vascular endothelial growth factor, which is an angiogenic factor that promotes vascular permeability. For example, certain compounds are tested for the ability to inhibit the tyrosine kinase activity associated with the VEGF receptors such as Flt and/or KDR and for their ability to inhibit angiogenesis and/or increased vascular permeability. Additionally, these compounds can be tested for the ability to inhibit the tyrosine kinase activity associated with Src and for their ability to inhibit angiogenesis and/or increased vascular permeability. These properties may be assessed, for example, using one or more of the procedures set out below:

Detailed Description Text - DETX (606):

This test measures the capacity of compounds to reduce the acute increase in uterine weight in rats which occurs in the first 4-6 hours following oestrogen stimulation. This early increase in uterine weight has long been known to be due to oedema caused by increased permeability of the uterine vasculature and recently Cullinan-Bove and Koos (Endocrinology, 1993,133:829-837) demonstrated a close temporal relationship with increased expression of VEGF mRNA in the uterus. It has been found that prior treatment of the rats with a neutralising monoclonal antibody to VEGF significantly reduces the acute increase in uterine weight, confirming that the increase in weight is substantially mediated by VEGF.

Claims Text - CLTX (39):

38. The composition of claim 37, wherein the composition further comprises an additional therapeutic agent and the therapeutic agent is an anticancer agent, an antiproliferative agent, an approved agent for the treatment of osteoporosis, or an approved agent for the treatment of disorders related to increased vascular permeability.

US-PAT-NO: 6693108

DOCUMENT-IDENTIFIER: US 6693108 B2

TITLE: Inhibitors of c-JUN N terminal kinases (JNK) and other protein kinases

DATE-ISSUED: February 17, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Green; Jeremy	Burlington	MA	N/A	N/A
Bemis; Guy	Arlington	MA	N/A	N/A
Grillot; Anne-Laure	Cambridge	MA	N/A	N/A
Ledeboer; Mark	Acton	MA	N/A	N/A
Salituro; Francesco G.	Marlboro	MA	N/A	N/A
Harrington; Edmund	South Boston	MA	N/A	N/A
Gao; Huai	Natick	MA	N/A	N/A
Baker; Christopher	Bedford	MA	N/A	N/A
Cao; Jingrong	Newton	MA	N/A	N/A
Hale; Michael	Bedford	MA	N/A	N/A

APPL-NO: 10/ 074177

DATE FILED: February 12, 2002

PARENT-CASE:

This application is a continuation and claims the benefit, under 35 U.S.C. .sctn.365(c) and .sctn.120, of co-pending PCT International Application PCT/US00/22445 filed Aug. 11, 2000. PCT International Application PCT/US00/22445 was published under PCT Article 21(2) in English and claims priority under 35 U.S.C .sctn.119 to U.S. Provisional Application Ser. No. 60/148,795 filed Aug. 13, 1999, U.S. Provisional Application Ser. No. 60/166,922 filed Nov. 22, 1999, and U.S. Provisional Application Ser. No. 60/211,517 filed Jun. 14, 2000, and the entire contents of each of these applications are hereby incorporated by reference.

US-CL-CURRENT: 514/275, 544/331

ABSTRACT:

The present invention provides compounds of formula I: ##STR1## where R.sup.1 is H, CONH.sub.2, T.sub.(n) --R, or T.sub.(n) --Ar.sup.2, n may be zero or one, and G, XYZ, and Q are as described below. These compounds are inhibitors of protein kinase, particularly inhibitors of JNK, a mammalian protein kinase involved cell proliferation, cell death and response to extracellular stimuli. The invention also relates to methods for producing these inhibitors. The invention also provides pharmaceutical compositions comprising the inhibitors of the invention and methods of utilizing those compositions in the treatment and prevention of various disorders.

11 Claims, 0 Drawing figures

Exemplary Claim Number: 1

---------- KWIC ---------

Brief Summary Text - BSTX (77):
In addition, JNK inhibitors of the instant invention may be capable of inhibiting the expression of inducible pro-inflammatory proteins. Therefore, other "JNK-mediated conditions" which may be treated by the compounds of this invention include edema, analgesia, fever and pain, such as neuromuscular pain, headache, cancer pain, dental pain and arthritis pain.

Brief Summary Text - BSTX (78):
The compounds of this invention are also useful as inhibitors of Src-family kinases, especially Src and Lck. For a general review of these kinases see Thomas and Brugge, Annu. Rev. Cell Dev. Biol. (1997) 13, 513; Lawrence and Niu, Pharmacol. Ther. (1998) 77, 81; Tatosyan and Mizenina, Biochemistry (Moscow) (2000) 65, 49. Accordingly, these compounds are useful for treating diseases or conditions that are known to be affected by the activity of one or more Src-family kinases. Such diseases or conditions include hypercalcemia, restenosis, hypercalcemia, osteoporosis, osteoarthritis, symptomatic treatment of bone metastasis, rheumatoid arthritis, inflammatory bowel disease, multiple sclerosis, psoriasis, lupus, graft vs. host disease, T-cell mediated hypersensitivity disease, Hashimoto's thyroiditis, Guillain-Barre syndrome, chronic obtructive pulmonary disorder, contact dermatitis, cancer, Paget's disease, asthma, ischemic or reperfusion injury, allergic disease, atopic dermatitis, and allergic rhinitis. Diseases that are affected by Src activity, in particular, include hypercalcemia, osteoporosis, osteoarthritis, cancer, symptomatic treatment of bone metastasis, and Paget's disease. Diseases that are affected by Lck activity, in particular, include autoimmune diseases, allergies, rheumatoid arthritis, and leukemia. Compounds of formula II-A and I-B wherein Ar.sup.2 is aryl are especially useful for treating diseases associated with the Src-family kinases, particularly Src or Lck.

Detailed Description Text - DETX (130):
Src Inhibition Assays

Detailed Description Text - DETX (131):
The compounds were assayed as inhibitors of full length recombinant human Src kinase (from Upstate Biotechnology, cat. no. 14-117) expressed and purified from baculo viral cells. Src kinase activity was monitored by following the incorporation of .sup.33 P from ATP into the tyrosine of a random poly Glu-Tyr polymer substrate of composition, Glu:Tyr 4:1 (Sigma, cat. no. P-0275). The following were the final concentrations of the assay components: 0.05 M HEPES, pH 7.6, 10 mM MgCl.sub.2, 2 mM DTT, 0.25 mg/ml BSA, 10 .mu.M ATP (1-2 .mu.Ci .sup.33 P-ATP per reaction), 5 mg/ml poly Glu-Tyr, and 1-2 units of recombinant human Src kinase. In a typical assay, all the reaction components with the exception of ATP were pre-mixed and aliquoted into assay plate wells. Inhibitors dissolved in DMSO were added to the wells to give a final DMSO concentration of 2.5%. The assay plate was incubated at 30.degree. C. for 10 min before initiating the reaction with .sup.33 P-ATP. After 20 min of reaction, the reactions were quenched with 150 .mu.l of 10% trichloroacetic acid (TCA) containing 20 mM Na.sub.3 PO.sub.4. The quenched samples were then transferred to a 96-well filter plate (Whatman, UNI-Filter GF/F Glass Fiber Filter, cat no. 7700-3310) installed on a filter plate vacuum manifold. Filter plates were washed four times with 10% TCA containing 20 mM Na.sub.3 PO.sub.4 and then 4 times with methanol. 200 .mu.l of scintillation fluid was then added to each well. The plates were sealed and the amount of radioactivity associated with the filters was quantified on a TopCount scintillation counter.

US-PAT-NO: 6685941

DOCUMENT-IDENTIFIER: US 6685941 B1

TITLE: Methods of treating autoimmune disease via CTLA-4Ig

DATE-ISSUED: February 3, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Thompson; Craig B.	Ann Arbor	MI	N/A	N/A
June; Carl H.	Rockville	MD	N/A	N/A

APPL-NO: 08/ 385194

DATE FILED: February 7, 1995

PARENT-CASE:

RELATED APPLICATIONS

This application is a continuation of U.S. Ser. No. 08/076,071, filed Jun. 10, 1993, now abandoned, which is a continuation-in-part of PCT/US93/03155, filed Apr. 6, 1993, which is a continuation of U.S. Ser. Nos. 07/864,805, 07/864,807, 07/864,866, all filed Apr. 7, 1992, and all abandoned, which are all continuation-in-parts of Ser. No. 07/275,433, filed Nov. 23, 1988, now abandoned, all of which are hereby incorporated by reference.

US-CL-CURRENT: 424/134.1, 424/192.1 , 514/2 , 514/8 , 514/885 , 530/350 , 530/387.3

ABSTRACT:

The method of immunotherapy of the present invention involves the regulation of the T cell immune response through the activation or suppression/inactivation of the CD28 pathway. Induction of activated T cell lymphokine production occurs upon stimulatory binding of the CD28 surface receptor molecule, even in the presence of conventional immunosuppressants. Inhibition of CD28 receptor binding to an appropriate stimulatory ligand or inactivation of the CD28 signal transduction pathway through other means down-regulates CD28-pathway related T cell lymphokine production and its resulting effects.

20 Claims, 33 Drawing figures

Exemplary Claim Number: 1

Number of Drawing Sheets: 25

---------- KWIC ---------

Detailed Description Text - DETX (33):

Studies with an inhibitor of the src family of tyrosine kinases, herbimycin A, and with tyrosine phosphatase, as described in Specific Example XI, further show that the functional effects of CD28 stimulation on lymphokine gene expression require the above-described protein tyrosine phosphorylation. The

data on tyrosine phosphorylation inhibitors thus demonstrate that inactivation of CD28-mediated signal transduction can also be used to down-regulate lymphokine production in accordance with the principles of the present invention.

Detailed Description Text - DETX (115):
An untreated animal and a CTLA-4Ig-treated animal were sacrificed for histological examination. Cardiac allografts were removed from an untreated animal (shown in FIG. 12A) and a CTLA-4Ig-treated animal (0.5 mg/day) (shown in FIG. 12B) four days after transplantation. Allografts were fixed in formalin, and tissue sections were stained with hematoxylin-eosin. (Original photography at 200.times.magnification.) The donor heart removed from the untreated animal showed histological findings of severe acute cellular rejection, including a prominent interstitial mononuclear cell infiltrate with edema formation, myocyte destruction, and infiltration of arteriolar walls. In contrast, the transplanted heart from the CTLA-4Ig-treated animal revealed only a mild lymphoid infiltrate. Frank myocyte necrosis and evidence of arteriolar involvement were absent. The native heart from each animal showed no histological abnormalities.

Detailed Description Text - DETX (132):
Herbimycin A prevents CD28-stimulated IL2 production. Previous studies have shown that three distinct biochemical signals, provided by phorbol esters, calcium ionophore, and ligation of the CD28 receptor with mAb, are required to cause optimal IL-2 secretion (see June, C. H. et al., J. Immunol., 143:153 (1989)). Cells cultured in the presence of PMA, ionomycin, or CD28 mAb alone produced no detectable IL-2 and, as previously reported in June, J. Immunol., (1989) supra, and Fraser, J. D. et al., Science (Wash., D.C.) 251:313 (1991), stimulation of the CD28 receptor strongly up-regulated IL-2 production of T cells stimulated with immobilized anti-CD2 mAb, PMA, or PMA plus ionomycin. To address the potential role of tyrosine kinases in CD28-triggered signaling, the effect of herbimycin A.sub.1 an inhibitor of the src family protein tyrosine kinases (see Uehara, Y. et al., Biochem. Biophys. Res. Commun., 163:803 (1989)), on the CD28-triggered enhancement of IL-2 production was investigated. T cells were cultured overnight in the absence (depicted as open bars in FIG. 15) or presence (depicted as filled bars in FIG. 15) of herbimycin A (1 .mu.M). The cells were then cultured for a further 24 h period in the presence of medium-immobilized anti-CD3 mAb (G19-4), PMA (3 ng/ml) (P), or PMA plus ionomycin (150 ng/ml) (P+I) in the presence or absence of soluble anti-CD28 mAb 9.3 (1 .mu.g/ml). Cell-free supernatant was collected, dialized to remove herbimycin A and serial dilutions were analyzed for IL-2 content by bioassay as described in June, J. Immunol., supra (1989). FIG. 15 shows the effect of herbimycin A on CD28-stimulated IL-2 production. The CD28 mAb mediated enhancement of IL-2 production in response to stimulation with immobilized anti-CD3, or PMA was nearly completely inhibited in the presence of herbimycin A. In contrast, cells cultured in PMA, ionomycin or 9.3 mAb only produced <10 U/ml of IL-2.

Detailed Description Text - DETX (143):
The brief temporal course of CD28 mAb-induced tyrosine phosphorylation suggested regulation by a phosphatase. To address the effects of phosphatases on CD28-mediated signal transduction, T cells were cultured overnight with PMA (5 ng/ml). 10.sup.7 cells were incubated for 10 m with media (control), biotinylated anti-CD45 mAb (9.4), anti-CD28 mAb (9.3), or both. Monoclonal antibodies were crosslinked with avidin at time 0. The reaction was terminated after 2 m. Immunoblot analysis with antiphosphotyrosine antibodies of detergent-soluble proteins was performed as previously described. CD28 crosslinking induced tyrosine phosphorylation on pp75 and pp100 that was completely prevented by CD45. Consistent with previous results described in

Samelson, L. E. et al., J. Immunol., 145:2448 (1990), crosslinking of CD45 alone caused increased tyrosine phosphorylation of a 120-135 kD substrate; this effect is also seen in CD28 plus CD45-treated cells. Thus, the above studies indicate that CD28-induced tyrosine phosphorylation is sensitive to an inhibitor of src family protein tyrosine kinases, and furthermore, that the CD45 protein tyrosine phosphatase can prevent CD28-induced protein tyrosine phosphorylation.

Other Reference Publication - OREF (84):
Uehara, Y. et al., "Irreversible Inhibition of V-SRC Tyrosine Kinase Activity by Herbimycin A and its Abrogation by Sulfhydryl Compounds," Biochem. Biophys. Res. Commun. 163:803-809 (1989).

US-PAT-NO: 6685938

DOCUMENT-IDENTIFIER: US 6685938 B1

TITLE: Methods and compositions useful for modulation of angiogenesis and vascular permeability using SRC or Yes tyrosine kinases

DATE-ISSUED: February 3, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Cheresh; David A.	Encinitas	CA	N/A	N/A
Eliceiri; Brian	Carlsbad	CA	N/A	N/A

APPL-NO: 09/ 470881

DATE FILED: December 22, 1999

PARENT-CASE:

GROSS-REFERENCE TO RELATED APPLICATIONS

This application is a continuation-in-part-of International Patent Application Number PCT/US99/11780, designating the United States of America filed May 28, 1999, which claims priority to United States Provisional Application for Patent Ser. No. 60/087,220 filed May 29, 1998.

US-CL-CURRENT: 424/94.5, 435/194 , 514/12

ABSTRACT:

The present invention describes methods for modulating vascular permeability (VP) in tissues using Src or modified Src protein, Yes protein or modified Yes protein, or mixtures thereof, and nucleic acids capable of expression such proteins. In particular, the invention describes methods for inhibiting VP using an inactive Src or Yes protein or a mixture thereof, or nucleic acids encoding therefor, or for potentiating VP using an active, Src or Yes protein or a mixture thereof, or nucleic acids encoding therefor. Related compositions and articles of manufacture are also disclosed.

16 Claims, 24 Drawing figures

Exemplary Claim Number: 1

Number of Drawing Sheets: 15

---------- KWIC ---------

Abstract Text - ABTX (1):

The present invention describes methods for modulating vascular permeability (VP) in tissues using Src or modified Src protein, Yes protein or modified Yes protein, or mixtures thereof, and nucleic acids capable of expression such proteins. In particular, the invention describes methods for inhibiting VP using an inactive Src or Yes protein or a mixture thereof, or nucleic acids encoding therefor, or for potentiating VP using an active, Src or Yes protein

or a mixture thereof, or nucleic acids encoding therefor. Related compositions and articles of manufacture are also disclosed.

TITLE - TI (1):
Methods and compositions useful for modulation of angiogenesis and vascular permeability using SRC or Yes tyrosine kinases

Brief Summary Text - BSTX (2):
The present invention relates generally to the field of medicine, and relates specifically to methods and compositions for modulating vascular permeability (VP).

Brief Summary Text - BSTX (6):
It has been proposed that inhibition of angiogenesis would be a useful therapy for restricting tumor growth. Inhibition of angiogenesis has been proposed by (1) inhibition of release of "angiogenic molecules" such as bFGF (basic fibroblast growth factor), (2) neutralization of angiogenic molecules, such as by use of anti-.beta.bFGF antibodies, (3) use of inhibitors of vitronectin receptor (.alpha..sub.v.beta..sub.3, and (4) inhibition of endothelial cell response to angiogenic stimuli. This latter strategy has received attention, Folkman et al., Cancer Biology, 3:89-96 (1992), have described several endothelial cell response inhibitors, including collagenase inhibitor, basement membrane turnover inhibitors, angiostatic steroids, fungal-derived angiogenesis inhibitors, platelet factor 4, thrombospondin, arthritis drugs such as D-penicillamine and gold thiomalate, vitamin D.sub.3 analogs, alpha-interferon, and the like that might be used to inhibit angiogenesis. For additional proposed inhibitors of angiogenesis, see Blood et al., Bioch. Biophys. Acta., 1032:89-118 (1990), Moses et al., Science, 248:1408-1410 (1990), Ingber et al., Lab. Invest., 59:44-51 (1988), and U.S. Pat. No. 5,092,885, U.S. Pat. No. 5,112,946, U.S. Pat. No. 5,192,744, U.S. Pat. No. 5,202,352, U.S. Pat. No. 5,753,230 and U.S. Pat. No. 5,766,591. None of the inhibitors of angiogenesis described in the foregoing references involve the Src proteins, however.

Brief Summary Text - BSTX (8):
The brain vasculature is characterized by a highly restrictive blood-brain barrier that prohibits small molecules from extravasating into the surrounding brain tissue. The nature of the blood-brain barrier in mammals has been of special concern with pharmacological studies, as many drugs are routinely prevented from passing from the vasculature to the brain tissues because of the highly restrictive blood-brain barrier. The present invention involves the unexpected discovery that VP, as measured by vascular leakage of blood, can be modulated by src or yes. Moreover, VP has been associated with angiogenesis and other pathologies. Inflammation induced increased vascular permeability is associated with edema and swelling.

Brief Summary Text - BSTX (10):
The present invention is directed to modulation of vascular permeability (VP) by tyrosine kinase Src, also referred to generically herein as Src, or the tyrosine kinase Yes, also referred to generically herein as Yes.

Brief Summary Text - BSTX (12):
In compositions which comprise active Src and Yes kinase proteins, the expected modulation is a potentiation or increase in vascular permeability of the blood vessels in a target tissue. Where the desired Src protein is an active kinase, a preferred Src is Src-A. Another preferred active Src protein is one in which the amino acid residue at position 527 of the Src protein is any amino acid residue except for tyrosine, serine or threonine. The preferred active Yes protein will have the kinase activity of wild-type human Yes, such

as that or the Yes-1 protein. Another preferred active Yes is one in which the kinase inactivating phosphorylation site of the Yes protein is mutated to abolish or minimize inactivating phosphorylation, similar to a mutation of amino acid residue 527 of Src to any amino acid residue except for tyrosine, serine or threonine.

Brief Summary Text - BSTX (13):
Where the composition comprises Src and Yes protein that are inactive kinase proteins, the expected modulation is an inhibition or decrease in vascular permeability of the blood vessels in the target tissue. When the desired Src protein is an inactive protein, a preferred Src is Src 251. A further preferred inactive Src is Src K295M. A preferred inactive Yes protein will have diminished kinase activity as compared with the wild-type protein.

Brief Summary Text - BSTX (15):
Where the modulation is a potentiation or increase in vascular permeability of the blood vessels in the target tissue, Src encoding nucleic acid will encode active forms of Src, and Yes encoding nucleic acids will encode active forms of Yes kinase proteins. Once transferred into the target host cell, the nucleic acids will be expressed by the host cell. A preferred Src encoding nucleic acid encodes active Src A protein. A further preferred Src encoding nucleic acid encodes a mutated active Src where the amino acid residue at position 527 of the expressed Src protein is any amino acid residue except for tyrosine, serine or threonine. A preferred Yes encoding nucleic acid will encode the wild-type protein, or a protein modified to abolish or inhibit the inactivating phosphorylation site of the Yes protein, in a similar manner as the Src position 527 mutation described.

Brief Summary Text - BSTX (16):
When the desired modulation is an inhibition or decrease in vascular permeability of the blood vessels in the target tissue, a preferred inactive Src encoding nucleic acid encodes Src 251 protein. A further preferred inactive Src encoding nucleic acid encodes inactive Src K295M. A preferred inactive Yes encoding nucleic acid will encode a protein that has diminished kinase activity.

Brief Summary Text - BSTX (25):
Where the Src or Yes protein is inactivated or inhibited, the modulation is an inhibition of VP. Where the Src or Yes protein is active or activated, the modulation is a potentiation of VP.

Brief Summary Text - BSTX (28):
The tissue to be treated can be any tissue in which modulation of VP is desirable. Therapeutic treatment is accomplished by contacting the target tissue with an effective amount of the desired modulating composition, and allowed sufficient time of contact for the protein or nucleic acid components of the pharmaceutical to enter the target tissue. For VP inhibition, it is useful to treat diseased tissue where deleterious vascular leaking is occurring. Exemplary tissues include inflamed tissue, tissues associated with stroke, myocardial infarction, or other blockage of normal flow, tissues undergoing restenosis, and the like tissues.

Brief Summary Text - BSTX (30):
A further aspect of the present invention are articles of manufacture which comprise packaging material and a pharmaceutical composition contained within said packaging material, wherein said pharmaceutical composition is capable of modulating vascular permeability in a tissue suffering from a disease condition, wherein said packaging material comprises a label which indicates that said pharmaceutical composition can be used for treating disease

conditions by modulating vascular permeability, and wherein said pharmaceutical composition comprises a therapeutically effective amount of tyrosine kinase protein Yes, in a pharmaceutically acceptable carrier. This embodiment encompasses Yes protein in active or inactive form, and also nucleic acids encoding for active or inactive Yes protein. Both retroviral and non-viral gene transfer/expression vectors can contain a nucleic acid segment encoding for Yes protein, either in active or inactive form, or both. When both active and inactive forms of a protein kinase gene are present, it is contemplated that the genes are under separate inducible promoter regulation to allow for alternative expression, as desired.

Brief Summary Text - BSTX (32):

A further aspect of the present invention are articles of manufacture which comprise a pharmaceutical composition wherein said pharmaceutical composition comprises a therapeutically effective VP modulating amount of an inactive tyrosine kinase protein Src and Yes protein, in a pharmaceutically acceptable carrier, where the desired modulation is an inactivation or inhibition of VP. A preferred inactive Src is Src 251 protein. Another preferred inactive Src protein is Src K295M.

Detailed Description Text - DETX (12):

The present invention relates generally to the discovery that VEGF induced VP is specifically mediated by the tyrosine kinase proteins Src and Yes, and that VP can be modulated by providing either active or inactive Src or Yes proteins for potentiating or inhibiting angiogenesis, respectively.

Detailed Description Text - DETX (13):

This discovery is important because of the role that vascular permeability plays in a variety of disease processes and in association with angiogenesis, the formation of new blood vessels. Where tissues associated with a disease condition require angiogenesis for tissue growth, it is desirable to inhibit angiogenesis and thereby inhibit the diseased tissue growth. Angiogenesis may be more effectively inhibited by simultaneously inhibiting VP. Where injured tissue requires angiogenesis for tissue growth and healing, it is desirable to potentiate or promote VP and thus angiogenesis, and thereby promote tissue healing and growth.

Detailed Description Text - DETX (18):

The present invention relates generally to the discovery that angiogenesis is mediated by the tyrosine kinase Src protein, and that angiogenesis can be modulated by providing either active or inactive Src proteins for potentiating or inhibiting angiogenesis, respectively.

Detailed Description Text - DETX (31):

An "inactive Src protein" refers to any of a variety of forms of Src protein which inhibit angiogenesis or VP. An "inactive Yes protein" refers to any of a variety of forms of Yes protein which inhibit VP. Assays to measure inhibition of VP increase are described herein, and are not to be construed as limiting. A Src protein is considered inactive if the level of angiogenesis is at least 10% lower, preferably 25% lower, and more preferably 50% lower than a control level where no exogenous Src is added to the assay system.

Detailed Description Text - DETX (82):

E. Methods For Modulation of Vascular Permeability (VP)

Detailed Description Text - DETX (83):

The invention provides for a method for the modulation of vascular permeability (VP) of the blood vessels in a tissue associated with a disease process or condition, and thereby effect events in the tissue which depend upon

VP. Generally, the method comprises administering to the tissue, associated with a disease process or condition, a composition comprising a VP-modulating amount of a Src or Yes protein, or mixture thereof, or nucleic acid vector expressing active or inactive Src or Yes, or both, according to the methods of this invention.

Detailed Description Text - DETX (87):
The dosage ranges for the administration of a Src or Yes protein depend upon the form of the protein, and its potency, as described further herein, and are amounts large enough to produce the desired effect in which VP and the disease symptoms mediated by VP are ameliorated. The dosage should not be so large as to cause adverse side effects, such as hyperviscosity syndromes, pulmonary edema, congestive heart failure, and the like. Generally, the dosage will vary with the age, condition, sex and extent of the disease in the patient and can be determined by one of skill in the art. The dosage can also be adjusted by the individual physician in the event of any complication.

Detailed Description Text - DETX (94):
Similarly, vascular permeability is an important component of angiogenesis, and in its own right associated with detrimental pathologies. For example, damage due to stroke induced vascular permeability triggers inflammation related damage.

Detailed Description Text - DETX (95):
Thus, methods which inhibit vascular permeability in a tissue associated with a disease condition ameliorates symptoms of the disease and, depending upon the disease, can contribute to cure of the disease. In one embodiment, the invention contemplates inhibition of vascular permeability, per se, in a tissue associated with a disease condition. The extent of vascular permeability in a tissue, and therefore the extent of inhibition achieved by the present methods, can be evaluated by a variety of methods.

Detailed Description Text - DETX (96):
Thus, in one related embodiment, a tissue to be treated is an inflamed tissue and the vascular permeability to be inhibited is due to VEGF mediated stimulation. In this class the method contemplates inhibition of VP in arthritic tissues, such as in a patient with chronic articular rheumatism, in immune or non-immune inflamed tissues, in psoriatic tissue and the like.

Detailed Description Text - DETX (99):
In a related embodiment, the invention contemplates the practice of the method in conjunction with other therapies such as conventional chemotherapy directed against solid tumors and for control of establishment of metastases. The administration of VP inhibitor is typically conducted during or after chemotherapy, although it is preferably to inhibit VP after a regimen of chemotherapy at times where the tumor tissue will be responding to the toxic assault by inducing VP to recover by the provision of a blood supply and nutrients to the tumor tissue. In addition, it is possible to administer the vascular permeability inhibition methods after surgery where solid tumors have been removed as a prophylaxis against metastases.

Detailed Description Text - DETX (100):
Insofar as the present methods apply to inhibition vascular permeability involved with metastases, the methods can also apply to inhibition of metastases as formation, and to regression of established tumors.

Detailed Description Text - DETX (101):
Restenosis is a process of smooth muscle cell (SMC) migration and proliferation into the tissue at the site of percutaneous transluminal coronary

angioplasty which hampers the success of angioplasty. The migration and proliferation of SMC's during restenosis can be considered a process of VP which is inhibited by the present methods. Therefore, the invention also contemplates inhibition of restenosis by inhibiting vascular permeability according to the present methods in a patient following angioplasty procedures. For inhibition of restenosis, the inactivated tyrosine kinase is typically administered after the angioplasty procedure because the coronary vessel wall is at risk of restenosis, typically for from about 2 to about 28 days, and more typically for about the first 14 days following the procedure.

Detailed Description Text - DETX (102):
The present method for inhibiting vascular permeability in a tissue associated with a disease condition, and therefore for also practicing the methods for treatment of vascular permeability-related diseases, comprises contacting a tissue in which increased vascular permeability is occurring, or is at risk for occurring, with a composition comprising a therapeutically effective amount of an inactivated Src and/or Yes protein or vector expressing the protein.

Detailed Description Text - DETX (104):
For example, manipulation of the permeability of the blood-brain barrier to modulate the access of drugs to the brain tissue is contemplated. An increase in vascular permeability of the blood-brain barrier will allow for drugs, that may normally not cross the barrier, to enter in to the brain tissues.

Detailed Description Text - DETX (116):
A therapeutic composition contains a vascular permeability-modulating amount of a Src and/or Yes protein of the present invention, or sufficient recombinant DNA expression vector to express an effective amount of Src and/or Yes protein, typically formulated to contain an amount of at least 0.1 weight percent of Src or Yes protein per weight of total therapeutic composition. A weight percent is a ratio by weight of Src or Yes protein to total composition. Thus, for example, 0.1 weight percent is 0.1 grams of Src or Yes protein per 100 grams of total composition. For DNA expression vectors, the amount administered depends on the properties of the expression vector, the tissue to be treated, and the like considerations.

Detailed Description Text - DETX (121):
The packaging material comprises a label which indicates the use of the pharmaceutical agent contained therein, e.g., for treating conditions assisted by the inhibition or potentiation of vascular permeability, and the like conditions disclosed herein. The label can further include instructions for use and related information as may be required for marketing. The packaging material can include container(s) for storage of the pharmaceutical agent.

Detailed Description Text - DETX (131):
Other mutations in Src are herein shown to have the opposite modulatory effect on VP, inhibiting VP instead of stimulating it. Such mutations are referred to as inactive Src mutations. Proteins having mutation that confer this inhibitory activity are also referred to as dominant negative Src proteins in that they inhibit VP, including that resulting from endogenous activity of Src as well as enhanced Src activity resulting from growth factor stimulation. Thus certain mutations of wild type c-src of the present invention can also function as a dominant negative with respect to their ability to block blood vessel growth and VP, and for example, therefore decrease VP in vivo.

Detailed Description Text - DETX (132):
Such preferred inhibitory c-Src protein includes the Src 251 in which only the first 251 amino acids of Src are expressed. This construct lacks the

entire kinase domain and is therefore referred to as "kinase dead" Src protein. A second construct is the Src (K295M) mutation in which the lysine amino acid residue 295 is mutated into a methionine. This point mutation in the kinase domain prevents ATP binding and also blocks kinase-dependent Src functions related to vascular cell and tumor cell signaling and proliferation.

Detailed Description Text - DETX (166):

The results of the assays described above indicate that both bFGF and VEGF treated CAMS in the presence of RCAS-GFP controls induced angiogenesis over the Src-mediated baseline angiogenesis seen with mock or untreated CAM preparations. The expressed dominant negative mutant Src 251 was effective at inhibiting VEGF-induced angiogenesis back to baseline levels while not effective at inhibiting bFGF-mediated angiogenesis. The photomicrographs shown in FIG. 8B pictorially confirm the data shown in FIG. 8A. Thus, retrovirally expressed Src 251 is an effective angiogenesis inhibitor, when angiogenesis is induced with VEGF.

Detailed Description Text - DETX (190):

To further analyze the role of Src in angiogenesis, a murine model was employed. In this case, angiogenesis was induced by subcutaneous injection of growth factor-depleted Matrigel supplemented with either bFGF (100 ng/ml) or VEGF (400 ng/ml) in athymic wehl(nu/nu) adult mice and analyzed after 5 days (Passaniti et al., 1992). Angiogenesis was quantitated by removing and homogenizing tissue, isolating the proteins., and immunoblotting with a VEGF receptor antibody (flk-1) (FIG. 13A) that is specific for endothelial cells. As observed in the chick, expression of the kinase-deleted Src 251 cDNA blocked VEGF-induced angiogenesis in this murine model while having no effect on bFGF-induced angiogenesis (FIG. 13B). To establish the role of endogenous Src in this model, tissues were infected with a retrovirus expressing Cak that inhibits endogenous Src activity by phosphorylation of the C-terminal regulatory site (Nada et al., 1991, Nature 361:68-72). Expression of Cak blocked VEGF-, but not bFGF-, induced angiogenesis (FIG. 13), confirming a role for endogenous Src activity in VEGF-mediated angiogenesis. Neovascularization of these virus-infected VEGF-stimulated tissues was confirmed by direct immunofluorescence with a FITC-conjugated anti-DC34 antibody (FIG. 13) or an anti-flk-1 antibody and quantitated by enumerating the number of positively stained CD34 blood vessels in each cryosection (FIG. 13C).

Detailed Description Text - DETX (194):

Continuing the results obtained with chicken and mouse angiogenesis models, a direct genetic approach was employed to examine intradermal VEGF-induced angiogenesis in src.sup.- /.sup.- mice. Also examined were effects on vascular permeability, since it was known that VEGF both initiates new blood vessel growth and can promote vascular permeability (Senger et al., 1983 Science 218:983-985; Ferrera and Davis-Smyth, 1997, Endocr.Rev. 16:4-25).

Detailed Description Text - DETX (197):

It was found that there were identical viral expression levels in src.sup.+ /.sup.- and src.sup.- /.sup.- as determined by X-gal staining of .beta.-galactosidase-adenovirus injected ears. In VEGF-injected src.sup.- /.sup.- ears, there was no significant decrease in angiogenesis as measured by counting branch points (p<0.05). However, surprisingly, the most apparent phenotype in these animals was the complete blockade of vascular leakage compared to the VEGF-injected src.sup.+ /.sup.- ears. Examination of ears injected with VEGF confirms the extent of the vascular leakage in src.sup.+ /.sup.- mice, that is essentially absent in the src.sup.- /.sup.- mice. The vascular leakage in these animals suggested that the VP activity, which has been associated with angiogenesis in vivo (Dvorak et al., 1995, Am.J.Pathol. 148:1029-1039), could be selectively disrupted in pp60.sup.c-src deficient

mice.

Detailed Description Text - DETX (201):
Vascular leakage of blood was localized to the VGEF-injected hemisphere in src.sup.+ /.sup.- mice, but there was a complete absence of vascular leakage in src.sup.- /.sup.- mice. This was also the case when examing the VP by measuring the accumulation of Evan's blue dye as detected by epifluoresence analysis of cryostat sections of these brains. Thus, VEGF compromises the blood-brain barrier in a manner that depends on active pp60.sup.c-src.

Detailed Description Text - DETX (203):
To further analyze and quantitate the effect of VEGF as a VP factor in src.sup.+ /.sup.- or src.sup.- /.sup.- mice, a Miles assay (Miles & Miles, 1952) was used to quantitatively measure the vascular permeability in the skin of these animals. VEGF was injected intradermally in src.sup.+ /.sup.- or src.sup.- /.sup.- mice that had received an intravenous systemic administration of Evan's blue dye. Within 15 min after injection of VEGF, there was a 3-fold increase in VP in src.sup.+ /.sup.- mice. However, in src.sup.- /.sup.- mice no detectable VP activity was observed. Dye elution of the injected skin patches were quantitated and compared with control saline and bFGF. bFGF or saline controls injected adjacent to the VEGF showed no significant increase in VP.

Detailed Description Text - DETX (205):
Vascular leakage/permeability is also known to occur during inflammation, which allows for the accumulation of serum-associated adhesive protein and inflammatory cells in tissues. In fact, inflammatory mediators themselves directly promote vascular leakage. Therefore, one such inflammatory mediator, allyl isothiocyanate, also known as mustard oil (Inoue et al., 1997, supra), was tested in src.sup.+ /.sup.- or src.sup.- /.sup.- mice for its capacity to produce VP. Unlike that observed in VEGF-stimulated src.sup.- /.sup.- animals, no decrease in the VP produced by the injection of the inflammatory mediator allyl isothiocyanate was detected. Thus, it can be concluded that Src plays a selective role in the VP activity induced with VEGF and does not influence VP associated with the inflammatory process.

Detailed Description Text - DETX (208):
The vascular permeability properties of VEGF in the skin of src.sup.+ /.sup.- (FIG. 14A, left panel) or src.sup.- /.sup.- (FIG. 14A, right panel) mice was determined by intradermal injection of saline or VEGF (400 ng) into mice that have been intravenously injected with Evan's blue dye. After 15 min, skin patches were photographed (scale bar, 1 mm). The stars indicate the injection sites. The regions surrounding the injection sites of VEGF, bFGF or saline were dissected, and the VP quantitatited by elution of the Evan's blue dye in formamide at 58.degree. C. for 24 hr, and the absorbance measured at 500 nm (FIG. 14B, left graph). The ability of an inflammation mediator (allyl isothiocyanate), known to induce inflammation related VP, was tested in src.sup.+ /.sup.- or src.sup.- /.sup.- mice (FIG. 14B, right).

Detailed Description Paragraph Table - DETL (1):
TABLE I Effect on Src/Mutation Src Function VP and Angiogenesis c-Src + active stimulates SrcA (T527F) + active stimulates Src527 (point) + active stimulates Src251 - inactive inhibits Src (truncate) - inactive inhibits Src (K295M) - inactive inhibits Src295 (point) - inactive inhibits

Claims Text - CLTX (8):
8. An article of manufacture comprising packaging material and a pharmaceutical composition contained within said packaging material, wherein said pharmaceutical composition is capable of modulating vascular permeability

in a tissue suffering from a disease condition, wherein said packaging material comprises a label which indicates that said pharmaceutical composition can be used for treating disease conditions by modulating vascular permeability, and wherein said pharmaceutical composition comprises a tyrosine kinase Src protein and Yes protein, in a pharmaceutically acceptable carrier and at least one of the Src protein and the Yes protein is an active kinase and at least one of the Src protein and the Yes protein is an inactive kinase.

Other Reference Publication - OREF (1):
van Bruggen, N. et al., 1999, "VEGF antagonism reduces edema formation and tissue damage after ischemia/reperfusion injury in the mouse brain" J. Clin. Invest. 104:1613-1620.

Other Reference Publication - OREF (2):
Hanke, J.H. et al., 1996, "Discovery of a novel, potent, and Src family-selective tyrosine kinase inhibitor" J. Biol. Chem. 271(2): 59: 6145-6152.

Other Reference Publication - OREF (3):
Moasser, M.M. et al., 1999, "Inhibition of Src kinases by a selective tyrosine kinase inhibitor causes mitotic arrest" Cancer Res. 59:6145-6152

Other Reference Publication - OREF (6):
Senger et al., 1983, "Tumor cells secrete a vascular permeability factor that promotes accumulation of ascities fluid" Science 219:983-985.

Other Reference Publication - OREF (7):
Maly et al., 2000, "Combinatorial target-guided ligand assembly: Identification of potent subtype-selective c-Src inhibitors" PNAS (USA) 97(6): 2419-2424.

US-PAT-NO: 6660744

DOCUMENT-IDENTIFIER: US 6660744 B1

TITLE: Pyrazolopyrimidines as therapeutic agents

DATE-ISSUED: December 9, 2003

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Hirst; Gavin C.	Marlborough	MA	N/A	N/A
Rafferty; Paul	Westborough	MA	N/A	N/A
Ritter; Kurt	Newton	MA	N/A	N/A
Calderwood; David	Framingham	MA	N/A	N/A
Wishart; Neil	Jefferson	MA	N/A	N/A
Arnold; Lee D.	Westborough	MA	N/A	N/A
Friedman; Michael M.	Newton	MA	N/A	N/A

APPL-NO: 09/ 663780

DATE FILED: September 15, 2000

PARENT-CASE:

RELATED APPLICATION

This application claims the benefit of United States Provisional Application No.: 60/154,620, filed Sep. 17, 1999, the entire teachings of which are incorporated herein by reference.

US-CL-CURRENT: 514/262.1, 514/210.21 , 544/262

ABSTRACT:

The present invention is directed to pyrazolopyrimidine derivatives which are useful as kinase inhibitors and as such are useful for affecting angiogenesis and diseases and conditions associated with angiogenesis.

86 Claims, 0 Drawing figures

Exemplary Claim Number: 1

---------- KWIC ---------

Brief Summary Text - BSTX (10):

As previously stated, recent evidence suggests that VEGF plays a role in the stimulation of both normal and pathological angiogenesis (Jakeman et al., Endocrinology 133: 848-859, 1993; Kolch et al., Breast Cancer Research and Treatment 36: 139-155, 1995; Ferrara et al., Endocrine Reviews 18(1); 4-25, 1997; Ferrara et al., Regulation of Angiogenesis (ed. L. D. Goldberg and E. M. Rosen), 209-232, 1997). In addition, VEGF has been implicated in the control and enhancement of vascular permeability (Connolly, et al., J. Biol. Chem. 264: 20017-20024, 1989; Brown et al., Regulation of Angiogenesis (ed. L. D. Goldberg and E. M. Rosen), 233-269, 1997).

Brief Summary Text - BSTX (13):

Placenta growth factor (PlGF) has an amino acid sequence that exhibits significant homology to the VEGF sequence (Park et al., J. Biol. Chem. 269:25646-54, 1994; Maglione et al. Oncogene 8:925-31, 1993). As with VEGF, different species of PlGF arise from alternative splicing of mRNA, and the protein exists in dimeric form (Park et al., supra). PlGF-1 and PlGF-2 bind to Flt-1 with high affinity, and PlGF-2 also avidly binds to neuropilin-1 (Migdal et al, J. Biol. Chem. 273 (35): 22272-22278), but neither binds to FLK-1/KDR (Park et al., supra). PlGF has been reported to potentiate both the vascular permeability and mitogenic effect of VEGF on endothelial cells when VEGF is present at low concentrations (purportedly due to heterodimer formation) (Park et al., supra).

Brief Summary Text - BSTX (17):
As for VEGF, VEGF-C and VEGF-D have been claimed to induce increases in vascular permeability in vivo in a Miles assay when injected into cutaneous tissue (PCT/US97/14696; WO98/07832, Witzenbichler et al., supra). The physiological role and significance of these ligands in modulating vascular hyperpermeability and endothelial responses in tissues where they are expressed remains uncertain.

Brief Summary Text - BSTX (19):
Based upon emerging discoveries of other homologs of VEGF and VEGFRs and the precedents for ligand and receptor heterodimerization, the actions of such VEGF homologs may involve formation of VEGF ligand heterodimers, and/or heterodimerization of receptors, or binding to a yet undiscovered VEGFR (Witzenbichler et al., supra). Also, recent reports suggest neuropilin-1 (Migdal et al, supra) or VEGFR-3/Flt-4 (Witzenbichler et al., supra), or receptors other than KDR/VEGFR-2 may be involved in the induction of vascular permeability (Stacker, S. A., Vitali, A., Domagala, T., Nice, E., and Wilks, A. F., "Angiogenesis and Cancer" Conference, Amer. Assoc. Cancer Res., January 1998, Orlando, Fla.; Williams, Diabetelogia 40: S118-120 (1997)).

Brief Summary Text - BSTX (26):
More recently, attempts have been made to identify small molecules which act as tyrosine kinase inhibitors. For example, bis monocyclic, bicyclic or heterocyclic aryl compounds (PCT WO 92/20642) and vinylene-azaindole derivatives (PCT WO 94/14808) have been described generally as tyrosine kinase inhibitors. Styryl compounds (U.S. Pat. No. 5,217,999), styryl-substituted pyridyl compounds (U.S. Pat. No. 5,302,606), certain quinazoline derivatives (EP Application No. 0 566 266 A1; Expert Opin. Ther. Pat. (1998), 8(4): 475-478), selenoindoles and selenides (PCTWO 94/03427), tricyclic polyhydroxylic compounds (PCT WO 92/21660) and benzylphosphonic acid compounds (PCT WO 91/15495) have been described as compounds for use as tyrosine kinase inhibitors for use in the treatment of cancer. Anilinocinnolines (PCT WO97/34876) and quinazoline derivative compounds (PCT WO97/22596; PCT WO97/42187) have been described as inhibitors of angiogenesis and vascular permeability.

Brief Summary Text - BSTX (27):
In addition, attempts have been made to identify small molecules which act as serine/threonine kinase inhibitors. For example, bis(indolylmaleimide) compounds have been described as inhibiting particular PKC serine/threonine kinase isoforms whose signal transducing function is associated with altered vascular permeability in VEGF-related diseases (PCT WO97/40830; PCT WO97/40831).

Brief Summary Text - BSTX (33):
Inhibitors of kinases involved in mediating or maintaining disease states represent novel therapies for these disorders. Examples of such kinases

include, but are not limited to: (1) inhibition of c-Src (Brickell, Critical Reviews in Oncogenesis, 3:401-406 (1992); Courtneidge, Seminars in Cancer Biology, 5:236-246 (1994), raf (Powis, Pharmacology & Therapeutics, 62:57-95 (1994)) and the cyclin-dependent kinases (CDKs) 1, 2 and 4 in cancer (Pines, Current Opinion in Cell Biology, 4:144-148 (1992); Lees, Current Opinion in Cell Biology, 7:773-780 (1995); Hunter and Pines, Cell, 79:573-582 (1994)), (2) inhibition of CDK2 or PDGF-R kinase in restenosis (Buchdunger et al., Proceedings of the National Academy of Science USA, 92:2258-2262 (1995)), (3) inhibition of CDK5 and GSK3 kinase in Alzheimers (Hosoi et al., Journal of Biochemistry (Tokyo), 117:741-749 (1995); Aplin et al., Journal of Neurochemistry, 67:699-707 (1996), (4) inhibition of c-Src kinase in osteoporosis (Tanaka et al., Nature, 383:528-531 (1996), (5) inhibition of GSK-3 kinase in type-2 diabetes (Borthwick et al., Biochemical & Biophysical Research Communications, 210:738-745 (1995), (6) inhibition of the p38 kinase in inflammation (Badger et al., The Journal of Pharmacology and Experimental Therapeutics, 279:1453-1461 (1996)), (7) inhibition of VEGF-R 1-3 and TIE-1 and -2 kinases in diseases which involve angiogenesis (Shawver et al., Drug Discovery Today, 2:50-63 (1997)), (8) inhibition of UL97 kinase in viral infections (He et al., Journal of Virology, 71:405-411 (1997)), (9) inhibition of CSF-1R kinase in bone and hematopoetic diseases (Myers et al., Bioorganic & Medicinal Chemistry Letters, 7:421-424 (1997), and (10) inhibition of Lck kinase in autoimmune diseases and transplant rejection (Myers et al., Bioorganic & Medicinal Chemistry Letters, 7:417-420(1997)).

Brief Summary Text - BSTX (35):

The identification of effective small compounds which specifically inhibit signal transduction and cellular proliferation by modulating the activity of receptor and non-receptor tyrosine and serine/threonine kinases to regulate and modulate abnormal or inappropriate cell proliferation, differentiation, or metabolism is therefore desirable. In particular, the identification of methods and compounds that specifically inhibit the function of a tyrosine kinase which is essential for antiogenic processes or the formation of vascular hyperpermeability leading to edema, ascites, effusions, exudates, and macromolecular extravasation and matrix deposition as well as associated disorders would be beneficial.

Brief Summary Text - BSTX (136):

In another aspect the present invention is directed to a method of treating a condition in a patient comprising administering a therapeutically effective amount of a compound of Formula (I) or a physiologically acceptable salt, prodrug or biologically active metabolites thereof to said patient, wherein said condition is an ocular condition, a cardiovascular condition, a cancer, Crow-Fukase (POEMS) syndrome, a diabetic condition, sickle cell anaemia, chronic inflammation, systemic lupus, glomerulonephritis, synovitis, inflammatory bowel disease, Crohn's disease, glomerulonephritis, rheumatoid arthritis, osteoarthritis, multiple sclerosis, graft rejection, Lyme disease, sepsis, von Hippel Lindau disease, pemphigoid, psoriasis, Paget's disease, polycystic kidney disease, fibrosis, sarcoidosis, cirrhosis, thyroiditis, hyperviscosity syndrome, Osler-Weber-Rendu disease, chronic occlusive pulmonary disease, asthma or edema following bums, trauma, radiation, stroke, hypoxia, ischemia, ovarian hyperstimulation syndrome, preeclampsia, menometrorrhagia, endometriosis, or infection by Herpes simplex, Herpes Zoster, human immunodeficiency virus, parapoxvirus, protozoa or toxoplasmosis.

Brief Summary Text - BSTX (137):

A preferred method is where the ocular condition is: ocular or macular edema, ocular neovascular disease, scleritis, radial keratotomy, uveitis, vitritis, myopia, optic pits, chronic retinal detachment, post-laser treatment complications, conjunctivitis, Stargardt's disease, Eales disease, retinopathy

or macular degeneration; the cardiovascular condition is: atherosclerosis, restenosis, ischemia/reperfusion injury, vascular occlusion or carotid obstructive disease; the cancer is: a solid tumor, a sarcoma, fibrosarcoma, osteoma, melanoma, retinoblastoma, a rhabdomyosarcoma, glioblastoma, neuroblastoma, teratocarcinoma, an hematopoietic malignancy, Kaposi's sarcoma, Hodgkin's disease, lymphoma, myeloma, leukemia or malignant ascites; and the diabetic condition is: insulin-dependent diabetes mellitus glaucoma, diabetic retinopathy or microangiopathy.

Brief Summary Text - BSTX (164):

Further, some of these compounds can be used as active agents against bums, chronic lung disease, stroke, polyps, anaphylaxis, chronic and allergic inflammation, delayed-type hypersensitivity, ovarian hyperstimulation syndrome, brain tumor-associated cerebral edema, high-altitude, trauma or hypoxia induced cerebral or pulmonary edema, ocular and macular edema, ascites, glomerulonephritis and other diseases where vascular hyperpermeability, effusions, exudates, protein extravasation, or edema is a manifestation of the disease. The compounds will also be useful in treating disorders in which protein extravasation leads to the deposition of fibrin and extracellular matrix, promoting stromal proliferation (e.g. keloid, fibrosis, cirrhosis and carpal tunnel syndrome). Increased VEGF production potentiates inflammatory processes such as monocyte recruitment and activation. The compounds of this invention will also be useful in treating inflammatory disorders such as inflammatory bowel disease (IBD) and Crohn's disease. ##STR52## ##STR53## ##STR54## ##STR55## ##STR56## ##STR57## ##STR58## ##STR59## ##STR60## ##STR61## ##STR62##

Brief Summary Text - BSTX (165):

VEGF's are unique in that they are the only angiogenic growth factors known to contribute to vascular hyperpermeability and the formation of edema. Indeed, vascular hyperpermeability and edema that is associated with the expression or administration of many other growth factors appears to be mediated via VEGF production. Inflammatory cytokines stimulate VEGF production. Hypoxia results in a marked upregulation of VEGF in numerous tissues, hence situations involving infarct, occlusion, ischemia, anemia, or circulatory impairment typically invoke VEGF/VPF mediated responses. Vascular hyperpermeability, associated edema, altered transendothelial exchange and macromolecular extravasation, which is often accompanied by diapedesis, can result in excessive matrix deposition, aberrant stromal proliferation, fibrosis, etc. Hence, VEGF-mediated hyperpermeability can significantly contribute to disorders with these etiologic features.

Brief Summary Text - BSTX (168):

The compounds of this invention have inhibitory activity against protein kinases. That is, these compounds modulate signal transduction by protein kinases. Compounds of this invention inhibit protein kinases from serine/threonine and tyrosine kinase classes. In particular, these compounds selectively inhibit the activity of the KDR/FLK-1/VEGFR-2 tyrosine kinases. Certain compounds of this invention also inhibit the activity of additional tyrosine kinases such as Flt-1/VEGFR-1, Flt-4, Tie-1, Tie-2, FGFR, PDGFR, IGF-1R, c-Met, Src-subfamily kinases such as Lck, Src, hck, fgr, fyn, yes, etc. Additionally, some compounds of this invention significantly inhibit serine/threonine kinases such as PKC, MAP kinases, erk, CDKs, Plk-1, or Raf-1 which play an essential role in cell proliferation and cell-cycle progression. The potency and specificity of the generic compounds of this invention towards a particular protein kinase can often be altered and optimized by variations in the nature, number and arrangement of the substituents (i.e., R.sub.1, R.sub.2, R.sub.3, A and ring 1) and conformational restrictions. In addition the metabolites of certain compounds may also possess significant protein kinase

inhibitory activity.

Brief Summary Text - BSTX (169):

The compounds of this invention, when administered to individuals in need of such compounds, inhibit vascular hyperpermeability and the formation of edema in these individuals. These compounds act, it is believed, by inhibiting the activity of KDR tyrosine kinase which is involved in the process of vascular hyperpermeability and edema formation. The KDR tyrosine kinase may also be referred to as FLK-1 tyrosine kinase, NYK tyrosine kinase or VEGFR-2 tyrosine kinase. KDR tyrosine kinase is activated when vascular endothelial cell growth factor (VEGF) or another activating ligand (such as VEGF-C, VEGF-D, VEGF-E or HIV Tat protein) binds to a KDR tyrosine kinase receptor which lies on the surface of vascular endothelial cells. Following such KDR tyrosine kinase activation, hyperpermeability of the blood vessels occurs and fluid moves from the blood stream past the blood vessel walls into the interstitial spaces, thereby forming an area of edema. Diapedesis also often accompanies this response. Similarly, excessive vascular hyperpermeability can disrupt normal molecular exchange across the endothelium in critical tissues and organs (e.g., lung and kidney), thereby causing macromolecular extravasation and deposition. Following this acute response to KDR stimulation which is believed to facilitate the subsequent angiogenic process, prolonged KDR tyrosine kinase stimulation results in the proliferation and chemotaxis of vascular endothelial cells and formation of new vessels. By inhibiting KDR tyrosine kinase activity, either by blocking the production of the activating ligand, by blocking the activating ligand binding to the KDR tyrosine kinase receptor, by preventing receptor dimerization and transphosphorylation, by inhibiting the enzyme activity of the KDR tyrosine kinase (inhibiting the phosphorylation function of the enzyme) or by some other mechanism that interrupts its downstream signaling (D. Mukhopedhyay et al., Cancer Res. 58:1278-1284 (1998) and references therein), hyperpermeability, as well as associated extravasation, subsequent edema formation and matrix deposition, and angiogenic responses, may be inhibited and minimized.

Brief Summary Text - BSTX (175):

The method of the present invention is useful in the treatment of protein kinase-mediated conditions, such as any of the conditions described above. In one embodiment, the protein kinase-mediated condition is characterized by undesired angiogenesis, edema, or stromal deposition. For example, the condition can be one or more more ulcers, such as ulcers caused by bacterial or fungal infections, Mooren ulcers and ulcerative colitis. The condition can also be due to a microbial infection, such as Lyme disease, sepsis, septic shock or infections by Herpes simplex, Herpes Zoster, human immunodeficincy virus, protozoa, toxoplasmosis or parapoxvirus; an angiogenic disorders, such as von Hippel Lindau disease, polycystic kidney disease, pemphigoid, Paget's disease and psoriasis; a reproductive condition, such as endometriosis, ovarian hyperstimulation syndrome, preeclampsia or menometrorrhagia; a fibrotic and edemic condition, such as sarcoidosis, fibrosis, cirrhosis, thyroiditis, hyperviscosity syndrome systemic, Osler-Weber-Rendu disease, chronic occlusive pulmonary disease, asthma, and edema following burns, trauma, radiation, stroke, hypoxia or ischemia; or an inflammatory/immunologic condition, such as systemic lupus, chronic inflammation, glomerulonephritis, synovitis, inflammatory bowel disease, Crohn's disease, rheumatoid arthritis, osteoarthritis, multiple sclerosis and graft rejection. Suitable protein kinase-mediated conditions also include sickle cell anaemia, osteoporosis, osteopetrosis, tumor-induced hypercalcemia and bone metastases. Additional protein kinase-mediated conditions which can be treated by the method of the present invention include ocular conditions such as ocular and macular edema, ocular neovascular disease, scleritis, radial keratotomy, uveitis, vitritis, myopia, optic pits, chronic retinal detachment, post-laser complications,

conjunctivitis, Stargardt's disease and Eales disease, in addition to retinopathy and macular degeneration.

Brief Summary Text - BSTX (181):

In many pathological conditions (for example, solid primary tumors and metastases, Kaposi's sarcoma, rheumatoid arthritis, blindness due to inappropriate ocular neovascularization, psoriasis and atherosclerosis) disease progression is contingent upon persistent angiogenesis. Polypeptide growth factors often produced by the disease tissue or associated inflammatory cells, and their corresponding endothelial cell specific receptor tyrosine kinases (e.g., KDR/VEGFR-2, Flt-1/VEGFR-1, Flt-4, Tie-2/Tek and Tie) are essential for the stimulation of endothelial cell growth, migration, organization, differentiation and the establishment of the requisite new functional vasculature. As a result of the vascular permeability factor activity of VEGF in mediating vascular hyperpermeability, VEGF-stimulation of a VEGFR kinase is also believed to play an important role in the formation of tumor ascites, cerebral and pulmonary edema, pleural and pericardial effusions, delayed-type hypersensitivity reactions, tissue edema and organ dysfunction following trauma, burns, ischemia, diabetic complications, endometriosis, adult respiratory distress syndrome (ARDS), post-cardiopulmonary bypass-related hypotension and hyperpermeability, and ocular edema leading to glaucoma or blindness due to inappropriate neovascularization. In addition to VEGF, recently identified VEGF-C and VEGF-D, and virally-encoded VEGF-E or HIV-Tat protein can also cause a vascular hyperpermeability response through the stimulation of a VEGFR kinase. KDRNEGFR-2 and/or Tie-2 are expressed also in a select population of hematopoietic stem cells. Certain members of this population are pluripotent in nature and can be stimulated with growth factors to differentiate into endothelial cells and participate in vasculogenetic angiogenic processes. For this reason these have been called Endothelial Progenitor Cells (EPCs) (J. Clin. Investig. 103: 1231-1236 (1999)). In some progenitors, Tie-2 may play a role in their recruitment, adhesion, regulation and differentiation (Blood, 4317-4326 (1997)). Certain agents according to formula I capable of blocking the kinase activity of endothelial cell specific kinases could therefore inhibit disease progression involving these situations.

Brief Summary Text - BSTX (183):

The compounds of formula I or a salt thereof or pharmaceutical compositions containing a therapeutically effective amount thereof may be used in the treatment of protein kinase-mediated conditions, such as benign and neoplastic proliferative diseases and disorders of the immune system, as described above. For example, such diseases include autoimmune diseases, such as rheumatoid arthritis, thyroiditis, type 1 diabetes, multiple sclerosis, sarcoidosis, inflammatory bowel disease, Crohn's disease, myasthenia gravis and systemic lupus erythematosus; psoriasis, organ transplant rejection (eg. kidney rejection, graft versus host disease), benign and neoplastic proliferative diseases, human cancers such as lung, breast, stomach, bladder, colon, pancreas, ovarian, prostate and rectal cancer and hematopoietic malignancies (leukemia and lymphoma), and diseases involving inappropriate vascularization for example diabetic retinopathy, retinopathy of prematurity, choroidal neovascularization due to age-related macular degeneration, and infantile hemangiomas in human beings. In addition, such inhibitors may be useful in the treatment of disorders involving VEGF mediated edema, ascites, effusions, and exudates, including for example macular edema, cerebral edema, acute lung injury and adult respiratory distress syndrome (ARDS).

Brief Summary Text - BSTX (190):

The compounds of this invention can be administered to a human patient by themselves or in pharmaceutical compositions where they are mixed with suitable carriers or excipient(s) at doses to treat or ameliorate vascular

hyperpermeability, edema and associated disorders. Mixtures of these compounds can also be administered to the patient as a simple mixture or in suitable formulated pharmaceutical compositions. A therapeutically effective dose further refers to that amount of the compound or compounds sufficient to result in the prevention or attenuation of inappropriate neovascularization, progression of hyperproliferative disorders, edema, VEGF-associated hyperpermeability and/or VEGF-related hypotension. Techniques for formulation and administration of the compounds of the instant application may be found in "Remington's Pharmaceutical Sciences," Mack Publishing Co., Easton, Pa., latest edition.

Brief Summary Text - BSTX (193):
Alternatively, one may administer the compound in a local rather than a systemic manner, for example, via injection of the compound directly into an edematous site, often in a depot or sustained release formulation.

Brief Summary Text - BSTX (233):
In the compositions of the present invention the active compound may, if desired, be associated with other compatible pharmacologically active ingredients. For example, the compounds of this invention can be administered in combination with one or more additional pharmaceutical agents that inhibit or prevent the production of VEGF or angiopoietins, attenuate intracellular responses to VEGF or angiopoietins, block intracellular signal transduction, inhibit vascular hyperpermeability, reduce inflammation, or inhibit or prevent the formation of edema or neovascularization. The compounds of the invention can be administered prior to, subsequent to or simultaneously with the additional pharmaceutical agent, whichever course of administration is appropriate. The additional pharmaceutical agents include but are not limited to anti-edemic steroids, NSAIDS, ras inhibitors, anti-TNF agents, anti-IL1 agents, antihistamines, PAF-antagonists, COX-1 inhibitors, COX-2 inhibitors, NO synthase inhibitors, Akt/PTB inhibitors, IGF-1R inhibitors, PKC inhibitors and PI3 kinase inhibitors. The compounds of the invention and the additional pharmaceutical agents act either additively or synergistically. Thus, the administration of such a combination of substances that inhibit angiogenesis, vascular hyperpermeability and/or inhibit the formation of edema can provide greater relief from the deletrious effects of a hyperproliferative disorder, angiogenesis, vascular hyperpermeability or edema than the administration of either substance alone. In the treatment of malignant disorders combinations with antiproliferative or cytotoxic chemotherapies or radiation are anticipated.

Brief Summary Text - BSTX (294):
In vivo Uterine Edema Model

Brief Summary Text - BSTX (295):
This assay measures the capacity of compounds to inhibit the acute increase in uterine weight in mice which occurs in the first few hours following estrogen stimulation. This early onset of uterine weight increase is known to be due to edema caused by increased permeability of uterine vasculature. Cullinan-Bove and Koss (Endocrinology (1993), 133:829-837) demonstrated a close temporal relationship of estrogen-stimulated uterine edema with increased expression of VEGF mRNA in the uterus. These results have been confirmed by the use of neutralizing monoclonal antibody to VEGF which significantly reduced the acute increase in uterine weight following estrogen stimulation (WO 97/42187). Hence, this system can serve as a model for in vivo inhibition of VEGF signalling and the associated hyperpermeability and edema. Materials: All hormones were purchased from Sigma (St. Louis, Mo.) or Cal Biochem (La Jolla, Calif.) as lyophilized powders and prepared according to supplier instructions. Vehicle components (DMSO, Cremaphor EL) were purchased from Sigma (St. Louis,

Mo.). Mice (Balb/c, 8-12 weeks old) were purchased from Taconic (Germantown, N.Y.) and housed in a pathogen-free animal facility in accordance with institutional Animal Care and Use Committee Guidelines.

Brief Summary Text - BSTX (298):
Results demonstrate that certain compounds of the present invention inhibit the formation of edema when administered systemically by various routes.

Claims Text - CLTX (59):
39. A method of treating a condition in a patient comprising administering a therapeutically effective amount of a compound of claim 1 or a physiologically acceptable salt, prodrug or biologically active metabolites thereof to said patient, wherein said condition is Crow-Fukase (POEMS) syndrome, a diabetic condition, sickle cell anaemia, systemic lupus, glomerulonephritis, synovitis, inflammatory bowel disease, Crohn's disease, glomerulonephritis, rheumatoid arthritis, osteoarthritis, multiple sclerosis, graft rejection, Lyme disease, sepsis, von Hippel Lindau disease, pemphigoid, psoriasis, Paget's disease, polycystic kidney disease, fibrosis, sarcoidosis, cirrhosis, thyroiditis, hyperviscosity syndrome, Osler-Weber-Rendu disease, chronic occlusive pulmonary disease, asthma or edema following burns, trauma, radiation, hypoxia, ischemia, ovarian hyperstimulation syndrome, preeclampsia, menometrorrhagia, endometriosis, or infection by Herpes simplex, Herpes Zoster, human immunodeficiency virus, parapoxvirus, protozoa or toxoplasmosis.

Claims Text - CLTX (60):
40. The method of claim 39 wherein the ocular condition is ocular or macular edema, ocular neovascular disease, scleritis, radial keratotomy, uveitis, vitritis, myopia, optic pits, chronic retinal detachment, post-laser treatment complications, conjunctivitis, Stargardt's disease, Eales disease, retinopathy or macular degeneration.

```
  * * * * * * * * * * * * * * STN  Columbus  * * * * * * * * * * * * * * * *

FILE 'HOME' ENTERED AT 14:32:03 ON 29 JUL 2004

=> fil .bec
COST IN U.S. DOLLARS                                    SINCE FILE      TOTAL
                                                           ENTRY    SESSION
FULL ESTIMATED COST                                         0.21       0.21

FILES 'MEDLINE, SCISEARCH, LIFESCI, BIOTECHDS, BIOSIS, EMBASE, HCAPLUS, NTIS,
       ESBIOBASE, BIOTECHNO, WPIDS' ENTERED AT 14:32:23 ON 29 JUL 2004
ALL COPYRIGHTS AND RESTRICTIONS APPLY.  SEE HELP USAGETERMS FOR DETAILS.

11 FILES IN THE FILE LIST

=> s vascula?(3a)(permeab? or leak?) or edema?
FILE 'MEDLINE'
        355316 VASCULA?
         94470 PERMEAB?
         38055 LEAK?
          7175 VASCULA?(3A)(PERMEAB? OR LEAK?)
         78034 EDEMA?
L1       83875 VASCULA?(3A)(PERMEAB? OR LEAK?) OR EDEMA?

FILE 'SCISEARCH'
        217561 VASCULA?
         98628 PERMEAB?
         48720 LEAK?
          7933 VASCULA?(3A)(PERMEAB? OR LEAK?)
         35735 EDEMA?
L2       42475 VASCULA?(3A)(PERMEAB? OR LEAK?) OR EDEMA?

FILE 'LIFESCI'
         23812 VASCULA?
         17293 PERMEAB?
          5424 LEAK?
          1174 VASCULA?(3A)(PERMEAB? OR LEAK?)
          4203 EDEMA?
L3        5217 VASCULA?(3A)(PERMEAB? OR LEAK?) OR EDEMA?

FILE 'BIOTECHDS'
          3446 VASCULA?
          3035 PERMEAB?
           924 LEAK?
           114 VASCULA?(3A)(PERMEAB? OR LEAK?)
           294 EDEMA?
L4          396 VASCULA?(3A)(PERMEAB? OR LEAK?) OR EDEMA?

FILE 'BIOSIS'
       2092201 VASCULA?
         92346 PERMEAB?
         33884 LEAK?
          9028 VASCULA?(3A)(PERMEAB? OR LEAK?)
         57214 EDEMA?
L5       64663 VASCULA?(3A)(PERMEAB? OR LEAK?) OR EDEMA?

FILE 'EMBASE'
        358943 VASCULA?
         79895 PERMEAB?
         35023 LEAK?
          6650 VASCULA?(3A)(PERMEAB? OR LEAK?)
         78596 EDEMA?
L6       83830 VASCULA?(3A)(PERMEAB? OR LEAK?) OR EDEMA?
```

```
FILE 'HCAPLUS'
        134179 VASCULA?
        201269 PERMEAB?
         80062 LEAK?
          6399 VASCULA?(3A)(PERMEAB? OR LEAK?)
         29324 EDEMA?
L7       34582 VASCULA?(3A)(PERMEAB? OR LEAK?) OR EDEMA?

FILE 'NTIS'
          2460 VASCULA?
         12790 PERMEAB?
         14905 LEAK?
            87 VASCULA?(3A)(PERMEAB? OR LEAK?)
           787 EDEMA?
L8          858 VASCULA?(3A)(PERMEAB? OR LEAK?) OR EDEMA?

FILE 'ESBIOBASE'
         66757 VASCULA?
         50321 PERMEAB?
          9368 LEAK?
          2137 VASCULA?(3A)(PERMEAB? OR LEAK?)
          6844 EDEMA?
L9        8679 VASCULA?(3A)(PERMEAB? OR LEAK?) OR EDEMA?

FILE 'BIOTECHNO'
         33459 VASCULA?
         18106 PERMEAB?
          4574 LEAK?
          1268 VASCULA?(3A)(PERMEAB? OR LEAK?)
          3864 EDEMA?
L10       4950 VASCULA?(3A)(PERMEAB? OR LEAK?) OR EDEMA?

FILE 'WPIDS'
         20521 VASCULA?
        106287 PERMEAB?
        137277 LEAK?
           455 VASCULA?(3A)(PERMEAB? OR LEAK?)
          2388 EDEMA?
L11       2780 VASCULA?(3A)(PERMEAB? OR LEAK?) OR EDEMA?

TOTAL FOR ALL FILES
L12     332305 VASCULA?(3A)(PERMEAB? OR LEAK?) OR EDEMA?

=> s src
FILE 'MEDLINE'
L13       14360 SRC

FILE 'SCISEARCH'
L14       13600 SRC

FILE 'LIFESCI'
L15        5412 SRC

FILE 'BIOTECHDS'
L16         247 SRC

FILE 'BIOSIS'
L17       14097 SRC

FILE 'EMBASE'
L18       10606 SRC

FILE 'HCAPLUS'
L19       14125 SRC
```

```
FILE 'NTIS'
L20        2011 SRC

FILE 'ESBIOBASE'
L21        7242 SRC

FILE 'BIOTECHNO'
L22        7046 SRC

FILE 'WPIDS'
L23         821 SRC

TOTAL FOR ALL FILES
L24       89567 SRC

=> s l12 and l24
FILE 'MEDLINE'
L25          34 L1 AND L13

FILE 'SCISEARCH'
L26          39 L2 AND L14

FILE 'LIFESCI'
L27           9 L3 AND L15

FILE 'BIOTECHDS'
L28           1 L4 AND L16

FILE 'BIOSIS'
L29          43 L5 AND L17

FILE 'EMBASE'
L30          24 L6 AND L18

FILE 'HCAPLUS'
L31          60 L7 AND L19

FILE 'NTIS'
L32           1 L8 AND L20

FILE 'ESBIOBASE'
L33          19 L9 AND L21

FILE 'BIOTECHNO'
L34           9 L10 AND L22

FILE 'WPIDS'
L35          31 L11 AND L23

TOTAL FOR ALL FILES
L36         270 L12 AND L24

=> s dup rem l36
MISSING OPERATOR REM L36
The search profile that was entered contains terms or
nested terms that are not separated by a logical operator.

=> dup rem l36
PROCESSING COMPLETED FOR L36
L37          123 DUP REM L36 (147 DUPLICATES REMOVED)

=> d 1-10
```

L37 ANSWER 1 OF 123 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
DUPLICATE 1
TI Methods and compositions useful for modulation of angiogenesis and **vascular permeability** using **SRC** or Yes tyrosine kinases.
SO Official Gazette of the United States Patent and Trademark Office Patents, (Feb 3 2004) Vol. 1279, No. 1. http://www.uspto.gov/web/menu/patdata.html. e-file.
ISSN: 0098-1133 (ISSN print).
AU Cheresh, David A. [Inventor, Reprint Author]; Eliceiri, Brian [Inventor]
AN 2004:130012 BIOSIS

L37 ANSWER 2 OF 123 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 2
TI Inhibition of **src** family kinases for the treatment of reperfusion injury related to revascularization
SO PCT Int. Appl., 62 pp.
CODEN: PIXXD2
IN Lorsordo, Douglas W.
AN 2004:331894 HCAPLUS
DN 140:350577

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2004032709	A2	20040422	WO 2003-US31430	20031003

PI
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

L37 ANSWER 3 OF 123 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 3
TI Preparation of vasculostatic agents and methods of use
SO PCT Int. Appl., 230 pp.
CODEN: PIXXD2
IN Wrasidlo, Wolfgang; Doukas, John; Royston, Ivor; Noronha, Glenn; Hood, John D.; Dneprovskaia, Elena; Gong, Xianchang; Splittgerber, Ute; Zhao, Ningning
AN 2004:308364 HCAPLUS
DN 140:321386

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2004030635	A2	20040415	WO 2003-US31721	20031003

PI
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

L37 ANSWER 4 OF 123 WPIDS COPYRIGHT 2004 THOMSON DERWENT on STN
TI New pyrrolotriazine derivatives useful for treatment of proliferative disease e.g. cancer, inflammation and autoimmune disease.
PI WO 2004013145 A1 20040212 (200420)* EN 71 C07D487-04
RW: AT BE BG CH CY CZ DE DK EA EE ES FI FR GB GH GM GR HU IE IT KE LS

LU MC MW MZ NL OA PT RO SD SE SI SK SL SZ TR TZ UG ZM ZW
W: AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CO CR CU CZ DE DK
DM DZ EC EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR
KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NI NO NZ OM PG PH
PL PT RO RU SC SD SE SG SK SL SY TJ TM TN TR TT TZ UA UG US UZ VC
VN YU ZA ZM ZW
US 2004063708 A1 20040401 (200425) A61K031-53
IN BHIDE, R S; BORZILLERI, R M

L37 ANSWER 5 OF 123 MEDLINE on STN DUPLICATE 4
TI **Src** blockade stabilizes a Flk/cadherin complex, reducing **edema** and tissue injury following myocardial infarction.
SO Journal of clinical investigation, (2004 Mar) 113 (6) 885-94.
Journal code: 7802877. ISSN: 0021-9738.
AU Weis Sara; Shintani Satoshi; Weber Alberto; Kirchmair Rudolf; Wood Malcolm; Cravens Adrianna; McSharry Heather; Iwakura Atsushi; Yoon Young-Sup; Himes Nathan; Burstein Deborah; Doukas John; Soll Richard; Losordo Douglas; Cheresh David
AN 2004172915 MEDLINE

L37 ANSWER 6 OF 123 MEDLINE on STN DUPLICATE 5
TI Angiogenic signal triggered by ischemic stress induces myocardial repair in rat during chronic infarction.
SO Journal of molecular and cellular cardiology, (2004 Apr) 36 (4) 547-59.
Journal code: 0262322. ISSN: 0022-2828.
AU Fukuda Shoji; Kaga Shigeaki; Sasaki Hiroaki; Zhan Lijun; Zhu Li; Otani Hajime; Kalfin Reni; Das Dipak K; Maulik Nilanjana
AN 2004186500 IN-PROCESS

L37 ANSWER 7 OF 123 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI Matrix-specific activation of **Src** and Rho initiates capillary morphogenesis of endothelial cells
SO FASEB JOURNAL, (MAR 2004) Vol. 18, No. 3, pp. 457-468.
Publisher: FEDERATION AMER SOC EXP BIOL, 9650 ROCKVILLE PIKE, BETHESDA, MD 20814-3998 USA.
ISSN: 0892-6638.
AU Liu Y Q; Senger D R (Reprint)
AN 2004:311780 SCISEARCH

L37 ANSWER 8 OF 123 MEDLINE on STN
TI Influenza virus inhibits ENaC and lung fluid clearance.
SO American journal of physiology. Lung cellular and molecular physiology, (2004 Aug) 287 (2) L366-73.
Journal code: 100901229. ISSN: 1040-0605.
AU Chen Xi-Juan; Seth Shaguna; Yue Gang; Kamat Pradip; Compans Richard W; Guidot David; Brown Lou Ann; Eaton Douglas C; Jain Lucky
AN 2004342287 IN-PROCESS

L37 ANSWER 9 OF 123 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
TI Multiple regulatory pathways of **vascular permeability** factor/**vascular** endothelial growth factor (VPF/VEGF) expression in tumors.
SO Seminars in Cancer Biology, (April 2004) Vol. 14, No. 2, pp. 123-130. print.
ISSN: 1044-579X.
AU Mukhopadhyay, Debabrata [Reprint Author]; Datta, Kaustubh
AN 2004:215589 BIOSIS

L37 ANSWER 10 OF 123 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 6
TI Pyrazolopyrimidine and furopyrimidine protein kinase inhibitors and their therapeutic use
SO PCT Int. Appl., 94 pp.
CODEN: PIXXD2
IN Hirst, Gavin C.; Arnold, Lee D.; Burchat, Andrew; Wishart, Neil;

Calderwood, David; Wada, Carol K.; Michaelides, Michael R.; Ji, Zhiqin; Muckey, Melanie

AN 2003:777596 HCAPLUS

DN 139:272922

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2003080064	A1	20031002	WO 2003-US8950	20030321
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
US 2003199525	A1	20031023	US 2002-103098	20020321

=> d 11-20

L37 ANSWER 11 OF 123 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 7

TI Preparation of phosphorus-substituted pyridopyrimidones as therapeutic agents

SO PCT Int. Appl., 164 pp.
CODEN: PIXXD2

IN Metcalf, Chester A., III; Shakespeare, William C.; Sawyer, Tomi K.; Wang, Yihan; Bohacek, Regine

AN 2003:5788 HCAPLUS

DN 138:56078

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2003000270	A1	20030103	WO 2002-US19605	20020621
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
US 2003100572	A1	20030529	US 2002-177520	20020621
EP 1408985	A1	20040421	EP 2002-739940	20020621
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				

L37 ANSWER 12 OF 123 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 8

TI Preparation of phosphorus-substituted quinolines as therapeutic agents

SO PCT Int. Appl., 146 pp.
CODEN: PIXXD2

IN Metcalf, Chester A., III; Shakespeare, William C.; Sawyer, Tomi K.; Wang, Yihan; Bohacek, Regine

AN 2003:5784 HCAPLUS

DN 138:56077

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2003000266	A1	20030103	WO 2002-US19535	20020621
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR,				

LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

US 2003114486	A1	20030619	US 2002-177500	20020621
US 6713462	B2	20040330		
EP 1408971	A1	20040421	EP 2002-739935	20020621

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR

L37 ANSWER 13 OF 123 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 9
TI Preparation of phosphorus-substituted idolinones as therapeutic agents
SO PCT Int. Appl., 230 pp.
CODEN: PIXXD2
IN Shakespeare, William C.; Sawyer, Tomi K.; Metcalf, Chester A., III; Wang, Yihan; Bohacek, Regine
AN 2003:5770 HCAPLUS
DN 138:56076

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2003000251	A1	20030103	WO 2002-US19769	20020621

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

US 2003130234	A1	20030710	US 2002-177472	20020621

L37 ANSWER 14 OF 123 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 10
TI Preparation of phosphorus-substituted quinazolines as therapeutic agents
SO PCT Int. Appl., 188 pp.
CODEN: PIXXD2
IN Wang, Yihan; Metcalf, Chester A., III; Shakespeare, William C.; Sawyer, Tomi K.; Bohacek, Regine
AN 2003:5719 HCAPLUS
DN 138:73379

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2003000188	A2	20030103	WO 2002-US19633	20020621
WO 2003000188	A3	20031231		

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

US 2003100573	A1	20030529	US 2002-177595	20020621
EP 1408980	A2	20040421	EP 2002-780884	20020621

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR

L37 ANSWER 15 OF 123 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 11

TI Preparation of phosphorus-substituted pyrazolo- and pyrrolopyrimidines as therapeutic agents
SO PCT Int. Appl., 165 pp.
CODEN: PIXXD2
IN Shakespeare, William C.; Sawyer, Tomi K.; Metcalf, Chester A., III; Wang, Yihan; Bohacek, Regine; Sundaramoorthi, Rajeswari
AN 2003:5718 HCAPLUS
DN 138:56075

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2003000187	A2	20030103	WO 2002-US19632	20020621

PI
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

US 2003114467	A1	20030619	US 2002-177563	20020621

L37 ANSWER 16 OF 123 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 12
TI Preparation of phosphorus-substituted phenylamino-pyrimidines as therapeutic agents
SO PCT Int. Appl., 203 pp.
CODEN: PIXXD2
IN Metcalf, Chester A., III; Shakespeare, William C.; Sawyer, Tomi K.; Wang, Yihan; Bohacek, Regine
AN 2003:5717 HCAPLUS
DN 138:56074

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2003000186	A2	20030103	WO 2002-US19631	20020621
WO 2003000186	A3	20030410		

PI
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

EP 1408978	A2	20040421	EP 2002-742236	20020621

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR

L37 ANSWER 17 OF 123 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 13
TI Preparation of pyrazolopyrimidine and furopyrimidine protein kinase inhibitors and their therapeutic use
SO U.S. Pat. Appl. Publ., 44 pp.
CODEN: USXXCO
IN Hirst, Gavin C.; Arnold, Lee D.; Burchat, Andrew; Wishart, Neil; Calderwood, David; Wada, Carol K.; Michaelides, Michael R.; Ji, Zhiqin; Muckey, Melanie
AN 2003:950055 HCAPLUS
DN 140:5065

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 2003225098	A1	20031204	US 2003-394965	20030321

PI

L37 ANSWER 18 OF 123 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Preparation of phosphorus-substituted pyridopyridimines as therapeutic agents
SO PCT Int. Appl., 151 pp.
CODEN: PIXXD2
IN Metcalf, Chester A., III; Shakespeare, William C.; Sawyer, Toni K.; Wang, Yihan; Bohacek, Regine
AN 2003:5665 HCAPLUS
DN 138:56073

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2003000011	A2	20030103	WO 2002-US19768	20020621
WO 2003000011	A3	20030320		

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, DM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

US 2003105115	A1	20030605	US 2002-176790	20020621

L37 ANSWER 19 OF 123 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Preparation of 4-aminopyrrolopyrimidines as protein kinase inhibitors
SO U.S. Pat. Appl. Publ., 93 pp., Cont.-in-part of U.S. 6,001,839.
CODEN: USXXCO
IN Calderwood, David; Arnold, Lee; Mazdiyasni, Hormoz; Hirst, Gavin C.; Deng, Bojuan B.; Johnston, David N.; Rafferty, Paul; Tometzki, Gerald B.; Twigger, Helen L.; Munschauer, Rainer
AN 2003:777394 HCAPLUS
DN 139:292260

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI US 2003187001	A1	20031002	US 1999-399083	19990917
US 6001839	A	19991214	US 1998-42702	19980317

L37 ANSWER 20 OF 123 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Preparation of pyrrolopyrimidines as tyrosine kinase inhibitors
SO U.S. Pat. Appl. Publ., 166 pp., Cont.-in-part of Appl. No. PCT/US99/21560.
CODEN: USXXCO
IN Hirst, Gavin C.; Calderwood, David; Munschauer, Rainer; Arnold, Lee D.; Johnston, David N.; Rafferty, Paul
AN 2003:633320 HCAPLUS
DN 139:180075

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI US 2003153752	A1	20030814	US 2000-537167	20000329
US 6713474	B2	20040330		
WO 2000017203	A1	20000330	WO 1999-US21560	19990917

W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

ZA 2001002204	A	20020318	ZA 2001-2204	20010316

=> log y

COST IN U.S. DOLLARS	SINCE FILE ENTRY	TOTAL SESSION
FULL ESTIMATED COST	45.30	45.51

STN INTERNATIONAL LOGOFF AT 14:40:22 ON 29 JUL 2004